EARLY ESTABLISHMENT AND PHOTOCHEMICAL EFFICIENCY OF ORTETS FROM DIVERSE AGROCLIMATES IN A COLD-PRONE ENVIRONMENT

Gitali Das*, Sandeep Kumar *, R. Krishnakumarand James Jacob****

*Rubber Research Institute of India, Regional Research Station, Jalpaiguri-735 225, West Bengal, India

**Rubber Research Institute of India, Kottayam-686 009, Kerala, India

Received: 08 February 2016 Accepted: 05 December 2016

Das,G.,Kumar,S.,Krishnakumar,R. and Jacob,J.(2016). Early establishment and photochemical efficiency of ortets from diverse agroclimates in a cold-prone environment. *Rubber Science*, **29**(3): 286-297.

Ortets selected from polyclonal seedling plantations raised in five different regional stations of RRII were planted at Nagrakata in sub-Himalayan West Bengal in order to evaluate their performance under cold stress. Early establishment and growth in the field as well as maximum (Fv/Fm) and effective (Φ PSII) quantum yield of PSII during post monsoon, pre-winter and post winter period were measured. Among the ortets tested, growth was better for the ortets collected from Guwahati. The survival of leaves at the top whorl was highest for the ortets RRSD 1 and lowest for RRSA 98. The magnitude of depression in Fv/Fm from post-monsoon to pre-winter was the lowest in RRSD 1. The study indicated that based on initial establishment and chlorphyll fluorescence parameters, RRSD 1 would be a highly potential cold tolerant ortet and RRSA 98 was found to be a highly susceptible ortet in the prevailing cold climate of sub-Himalayan West Bengal.

Key words: Cold stress, *Hevea brasiliensis*, Ortet, Photosynthetic efficiency, Cold tolerance

INTRODUCTION

Adaptation of *Hevea*, a tropical plant, to cold climate was well established (Alam *et al*., 2002; Raj *et al*., 2005; *Das et al*., 2013 b). Some plants being well adapted to tropical climate would also show tolerance to chilling stress (Mai *et al*., 2010). Chilling stress leads to alterations in metabolic processes, decrease in enzymatic activities, reduction of photosynthetic capacity and changes in membrane permeability (Alam *et al*., 2005; Sevillano *et al*., 2009; Mai *et al*., 2009; Jing *et al*., 2010; Jacob, 2013). It was observed that tropical and subtropical plants when grown under chilling temperature showed adverse effect (Allen and Ort, 2001). *Hevea* polycross seedling progeny, when grown under lower Brahmaputra valley of Assam (Kamrup) and Terai zone of West Bengal (Jalpaiguri) over years showed variation in adaptation potential (Das *et al*., 2013a). On the basis of performance of polycross seedling trees raised in various regional research stations of RRII, selections were made and the superior performers (ortets) from Nagrakata, Dapchari, Agartala, Tura and Guwahati, were procured and grown in sub-Himalayan climatic condition. The weather pattern of four stations of North East India compared to that of Dapchari, Maharashtra are quite

Correspondence: Gitali Das (Email: gitalidas@rubberboard.org.in)

Table 1. **Geographical location and the weather parameters of the location**

Place of ortets selected	Geographical position	Weather parameters				
		T_{max}(^{0}C)	T_{min}(^{0}C)	SS (hr/day)	Total rain (mm/yr.)	No. of rainy days/yr
Ganolgre, Tura,	Latitude 25^{0}342 N	29.0	16.4	5.9	2460	90
Meghalaya	Longitude 90^{0}142 E	±	±	±	±	±
	Altitude 600 m MSL	3.2	5.6	0.4	768	11.0
Nagrakata, Jalpaiguri	Latitude 26^{0}512 N	29.4	17.3	5.5	3799	138
West Bengal	Longitude 88^{0}572 E	±	±	±	±	±
	Altitude 69 m MSL	4.3	6.9	0.5	560	13.0
Sarutari, Sonapur,	Latitude 26^{0}032 N	29.9	18.8	5.5	1412	92
Kamrup, Assam	Longitude 91^{0}532 E	±	±	±	±	±
	Altitude 125 m MSL	4.0	5.4	0.6	336	19.0
Mohanpur, Agartala,	Latitude 23^{0}57 2 N	30.6	20.1	6.4	1969	112
Tripura	Longitude 91^{0}21 2 E	±	±	±	±	±
	Altitude 35 m MSL	3.4	5.8	0.7	388	12.0
Dapchari, Thane,	Latitude 20^{0}05′2 N	33.2	20.7	7.5	2575	108
Maharashtra	Longitude 72^{0}54 2 E	±	±	±	±	±
	Altitude 60 m MSL	3.6	4.8	0.7	579	34.0

different in geographical terms as well as in terms of the prevailing weather parameters like temperature, sunshine duration and precipitation (Table 1). The annual mean maximum (T_{max}) temperature in Tura (Meghalaya), Nagrakata (Jalpaiguri, West Bengal), Sarutari (Kamrup, Assam) and Mohanpur (Agartala, Tripura) are similar but it is higher in Dapchari (Maharashtra). In Dapchari, the mean minimum temperature (T_{min}) was higher than that of Tura and Nagrakata. Compared to North East India, sunshine hours (SS) at Dapchari is more, with optimum, but, less distributed precipitation, while it is very high at Nagrakata. The ±SE value for the number of rainy days indicated that the range is very wide at Dapchari compared to other stations. Therefore, it is highly unstable/unpredictable precipitation with high T_{max} and more SS that made the weather of Dapchari quite different from the other stations.

The selected ortets were cultivated under widely different environment assuming that influence of the growing environment may be reflected in their performance on the basis of which potential smart ortet(s) would be screened for the region.

MATERIALS AND METHODS

The climate of Nagrakata where the *Hevea* ortets were grown in field condition was different from that of its traditional belt in terms of heavy rainfall, severe wind and severe low winter temperature. The degree of cold stress experienced by the plants after four months of field planting is depicted in Figure 1. The average T_{max} and T_{min} in January 2013 was 23.6^{o}C and 6.9^{o}C. Severe cold due to T_{min} d” 5^{o}C for five consecutive days was

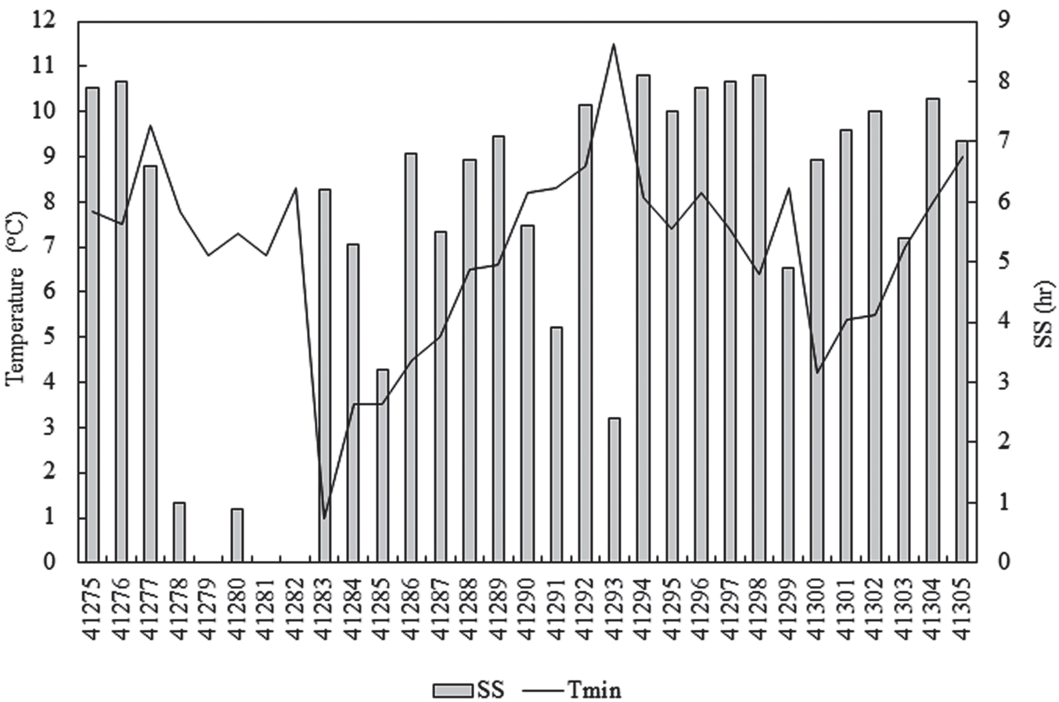

Fig. 1. Pattern of low winter temperature (T_{min}) and sunshine duration (hr) experienced by the plants after four months of field planting

observed from 9th (1.0°C) to 13th (5°C) January with sunshine duration ranging from 3.2 hr to 6.8 hr.

Sixteen ortets from different regions *viz.* Tura, Meghalaya (RRST), Agartala, Tripura (RRSA), Guwahati, Assam (RRSG), Nagrakata, West Bengal (RRSN) and Daphchari, Maharashtra (RRSD) were vegetatively multiplied (Table 1) along with two check clones RRIM 600 and RRII 429. The ortets from different regions were bud grafted and grafted stumps were maintained in polybag nursery till low winter temperature subsided. Polybag plants with two to three whorls of leaves were planted in the field with three replications having seven plants in each replication. Girth was measured at 30 cm height from the bud union during 13th and 22nd month of planting and girth increment between these two measurements was calculated for one year. Cold injury during January (winter period) in terms of leaf retention on the top most whorl and survival of the whole plant was assessed and expressed on percentage basis. Chlorophyll data recording was conducted from seven plants, two leaves from each plant and from eight spots from each leaf. Extent of chlorophyll degradation during pre-winter (last week of December) after four month of field establishment was assessed using chlorophyll content meter-CCM 200 (Opti-Sciences, USA). Total chlorophyll content, chlorophyll *a* and chlorophyll *b* of this period were calculated from chlorophyll content index (CCI) data following the regression equation by Nair and Jacob (2011). After one year of field planting, level of chlorophyll degradation during post-monsoon (September) and winter (January) was assessed using SPAD-502 Plus chlorophyll meter (Spectrum Technologies Inc.). The efficiency of excitation energy in dark adapted condition and photosynthetic efficiency of PS II reaction centres under light exposed condition during post-monsoon (October), pre-winter (December) and post-winter (May) period was measured in mature leaves attached to the plant by portable pulse modulated Fluorescence Monitoring System (FMS2, Hansatech Instruments Ltd., Norfolk, UK). Intact leaves from second whorl from the top of nine plants each were dark-adapted with leaf clips for 30 min to allow relaxation of fluorescence quenching associated with thylakoid membrane energization (Krause *et al*., 1983). Minimal fluorescence (Fo) and maximal fluorescence (Fm) were obtained by imposing a one second saturating flash to reduce all the PS II reaction centres. The maximum potential photochemical efficiency of PS II was expressed as the ratio Fv/Fm = (Fm " Fo)/Fm). Effective PS II quantum yield (Φ PSII) under light condition was calculated (Schreiber *et al.*, 1998) as follows: Φ PSII = (Fm′ – Ft / Fm′). The degree of photoinhibition / quantum photoefficiency was evaluated by the reduction in the value of Fv/Fm and Φ PSII. Chlorophyll and photo-efficiency data was recorded from nine plants per clone and minimum of two leaves per plant. Meteorological data of the stations were collected from the meteorological observatories of the respective stations. All the data were subjected to simple statistical analysis using ANOVA.

RESULTS AND DISCUSSION

The planting material of all the ortets / clones were of good quality and there was no disease attack just after field planting. Among the tested ortets, all except one (RRST 39) showed 100 per cent survival after three and half months of field planting and before the onset of subsequent cold stress (Table 2). RRST 39 showed only 81 per cent survival, indicating that probably the sub-Himalayan environment was not conducive for this ortets which was collected from Tura, Meghalaya. The field survival after cold stress in 17 month old plants was maximum (100%) for the clones RRSD 1, RRSA 585, RRSG 1, RRSG 3, RRSG 9, RRST 37 and the check clone RRIM 600, followed by RRSD 36, RRSN 47, RRSN 69 and RRII 429 (95%). The

Table 2. **Preliminary adaptation potential of ortets in field condition under the agroclimate of Sub-Himalayan West Bengal**

Name of Ortet	Survival rate in field before onset of cold stress (%)	Survival rate in field after cold stress (%)	Survival rate of leaves at the top most whorl after cold stress (%)
RRSD 1	100	100	72
RRSD 34	100	90	80
RRSD 35	100	86	46
RRSD 36	100	95	38
RRSN 1	100	86	64
RRSN 47	100	95	79
RRSN 69	100	95	90
RRSA 98	100	67	8
RRSA 315	100	86	30
RRSA 585	100	100	100
RRSG 1	100	100	100
RRSG 3	100	100	100
RRSG 9	100	100	55
RRST 24	100	90	70
RRST 37	100	100	74
RRST 39	81	86	66
RRIM 600	100	100	100
RRII 429	100	95	79
CD ($P \geq 0.05$)	3.24	6.20	18.90

lowest field survival was for clone RRSA 98 (67%). Reduction in growth at low temperature (monthly mean temperature below 20^{0}C) was reported earlier in *Hevea* (Jiang, 1988). Also,low temperature and high irradiance showed significant influence on leaf area ratio in *Hevea* (Ray *et al.*, 2004).

Effect of cold shock was reflected on the survival rate of leaves at the top most whorl (Table 2). The survival rate of leaves at the top most whorl also varied with the ortets indicating the differences in their cold tolerance capacity. The highest leaf survival was recorded for the clones, RRST 585, RRSG 1, RRSG 3 and check clone RRIM 600 (100%) followed by RRSN 69 (90%), RRSD 34 (80%), RRSN 47 (79%), RRST 37 (74%), RRSD 1 (72%) and RRII 429 (79%). The lowest survival was for RRSA 98 (8%). For all other clones the survival value ranged from 70-30%. In Birch, shoot apex acted as an indicator of cold response by changing its intrinsic activity under stress in the form of dormant bud; it was found to regain its activity after receiving favourable environment (Rinne *et al.*, 2001).

Table 3. **Early growth performance of ortets in field condition**

Name of ortets	Girth* (cm)		
	13 months of field planting	22 months of field planting	GI (cm)
RRSD 1	2.7	11.8	9.14
RRSD 34	2.9	12.4	9.42
RRSD 35	3.4	12.7	9.26
RRSD 36	1.7	9.0	7.32
RRSN 1	2.9	11.4	8.6
RRSN 47	3.1	12.0	8.9
RRSN 69	3.6	13.2	9.6
RRSA 98	2.6	9.5	6.9
RRSA 315	3.2	12.0	8.8
RRSA 585	4.0	13.8	9.8
RRSG 1	5.0	5.5	10.5
RRSG 3	4.4	14.3	9.9
RRSG 9	3.5	14.4	10.9
RRST 24	2.9	11.8	8.9
RRST 37	3.4	13.7	10.3
RRST 39	2.3	10.5	8.2
RRIM 600	4.1	13.7	9.6
RRII 429	3.9	13.7	9.8
CD (P≥0.05)	0.8	1.0	

*at 30 cm height

In general, young plants are more vulnerable to extreme climatic conditions than grown up mature plants in *Hevea* at different climatic regions of India (Jacob *et al*., 1999). In the present study also the effect of prevailing weather of the sub-Himalayan West Bengal on the establishment and growth of the ortets brought from different locations was reflected at the early establishment stage itself. Considering the survival characters in the early stages of growth (Table 2), RRSD 1, RRSA 585, RRST 37 and all ortets of RRSG were found superior to the rest of the ortets. RRSA 98 was found to be the least preferred one for the region among the tested ortets based on the survival character.

The early growth performance in terms of girth of the ortets showed that highest growth was achieved by the ortets of group RRSG at 13 month after field planting as well as after 22 month (Table 3) followed by the check clones. The girth at 30 cm after 13 months of planting was highest for the clones RRSG1 (5.0 cm) followed by RRSG 3 (4.4 cm) and the check clone RRIM 600 (4.1 cm). The lowest girth was shown by RRSD 36. At 22 months of field planting also, the highest girth was recorded by RRSG 1 (15.5 cm) followed by RRSG 3 (14.3 cm) and RRSG 9 (14.4 cm). Girth of RRSG 3, RRSA 585, RRSN 69, RRSG 9, RRST 37 and RRSD 35 after 13 month of planting and RRSG 3.

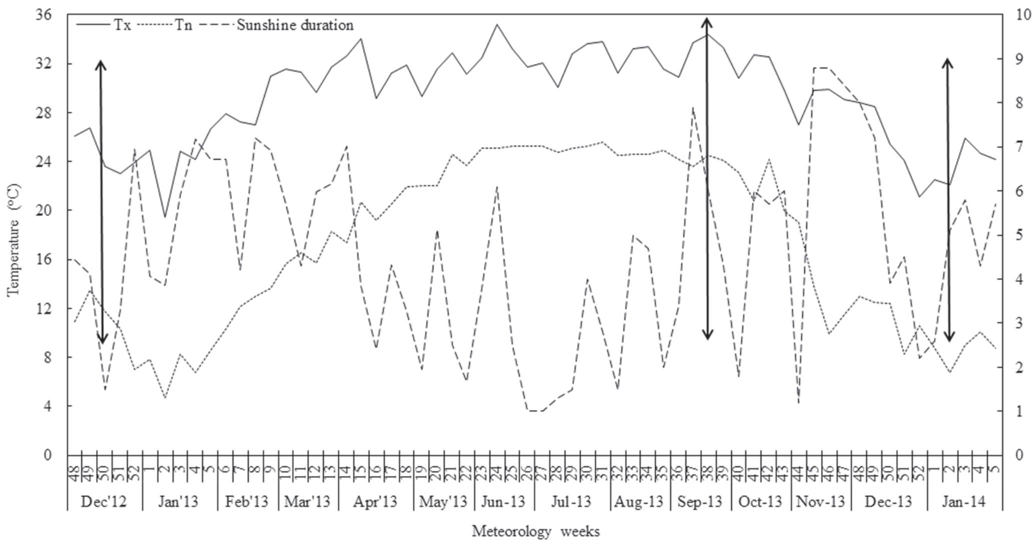

Fig. 2. Pattern of weather attributes during chlorophyll content measurement period

RRSG 9, RRSA 585 and RRST 37 after 22 month of field planting were on par with that of the check clones. The lowest girth was observed in RRSD 36 and RRSA 98 at 13 months of planting as well as after 22 months.

The difference between girth at 13^{th} month and at 22^{nd} month of growth (Table 3) showed that it was higher in RRSG 9 and RRSG 1 than that of the check clones; difference was low in RRSA 98 followed by RRSD 36 and RRST 39. The ortet RRSG 1, RRSG 3 and RRSG 9 showed better girth than other ortets during early establishment stage.

The comparative study on chlorophyll content at early establishment stage (four months after field planting), showed that the total chlorophyll content in RRSN 69 was significantly higher than the check clone RRIM 600 but at par with RRII 429 (Fig. 3). The second highest total chlorophyll content was in RRSA 315 and RRSD 34 followed by RRSG 9 and RRSG 1. The lowest chlorophyll content was observed in RRSD 1 and RRSA 585. The chlorophyll a/b ratio was significantly higher in RRSD 1 and RRSA 585 than the check clones RRIM 600 and RRII 429. Adjustment of chlorophyll a/b ratio was found to be apparently a good indicator of acclimatization at high light condition and low nitrogen availability (Kitajima and Hogan, 2003). Thus, high chlorophyll a/b ratio in RRSD 1 and RRSA 585 depicted that they were appreciably acclimatized to severe low winter temperature during the initial establishment phase compared to that of the other ortets / clones. Reduction in Chlorophyll a/b ratio due to cold was also reported in *Tamarix chinensis* seedling (Joo and Lee, 2011) which may result in decrease in photosynthetic efficiency (Renaut *et al.*, 2005).

The total chlorophyll in RRSN 69 was significantly higher than the check clone RRIM 600 (Fig. 4) during the following post-monsoon period (weekly mean T_{max} and T_{min} 34.4°C and 24.5°C, respectively with 6.0 hr SS - Fig. 2). A clear trend of reduction in chlorophyll content due to cold period was observed (when weekly mean T_{max}, T_{min} and

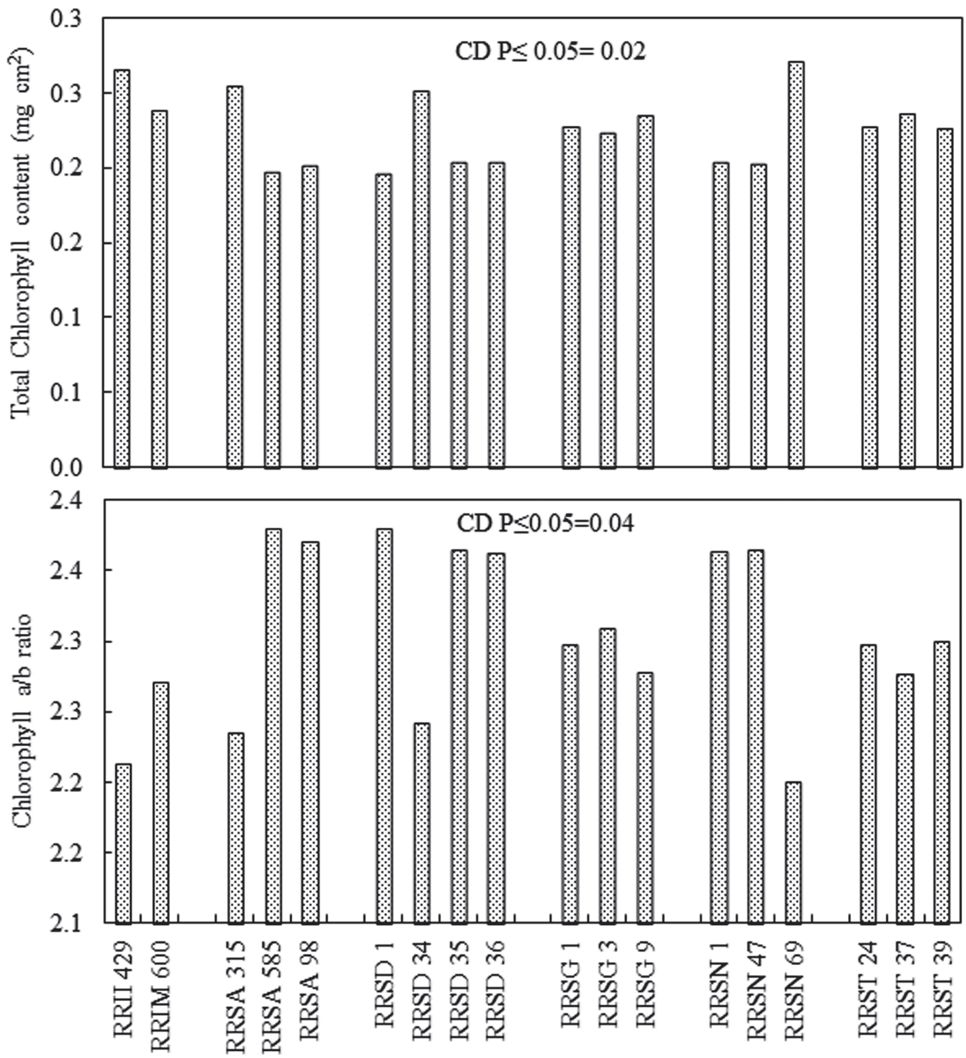

Fig. 3. Total chlorophyll content and chlorophyll a/b ratio of ortets in field condition during pre-winter after four month of field planting

SS as 25.9°C, 9°C and 5.8 hr, respectively during winter data recording time). The reduction in total chlorophyll content due to severe cold was significantly higher in RRSN 47 in comparison to that of the two check clones RRIM 600 and RRII 429 followed by RRSN 69, RRSD 36 and RRSN 1. Low temperature effect on chlorophyll was negligible in RRSA 585, RRST 39 and RRSD 35. Similar to our observations, reduction in total chlorophyll content due to cold stress has been reported in *Tamarix chinensis* (Joo and Lee, 2011).

The effect of cold stress was reflected on chlorophyll fluorescence parameters also. The PS II photochemistry was

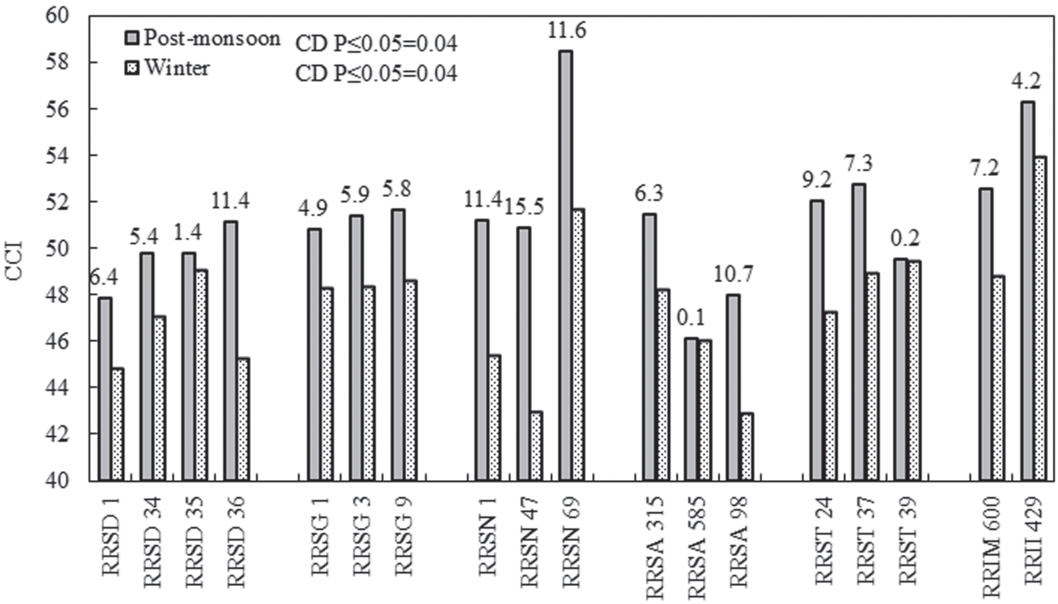

Fig. 4. Effect of severe low winter temperature on chlorophyll content in different ortets under field condition

measured under dark adapted (Fv/Fm) and light exposed (Φ PSII) conditions during post-monsoon period with T_{max}, T_{min} and SS as 31.3°C, 19.4°C and 8.0 hr., respectively. It was measured during pre-winter period also with T_{max}, T_{min} and SS as 23.9°C, 7.1°C and 6.9 hr., respectively and during post-winter period with T_{max}, T_{min} and SS at 32.9°C, 24.6°C and 2.5 hr., respectively (Fig. 5). The extent of depression in Fv/Fm

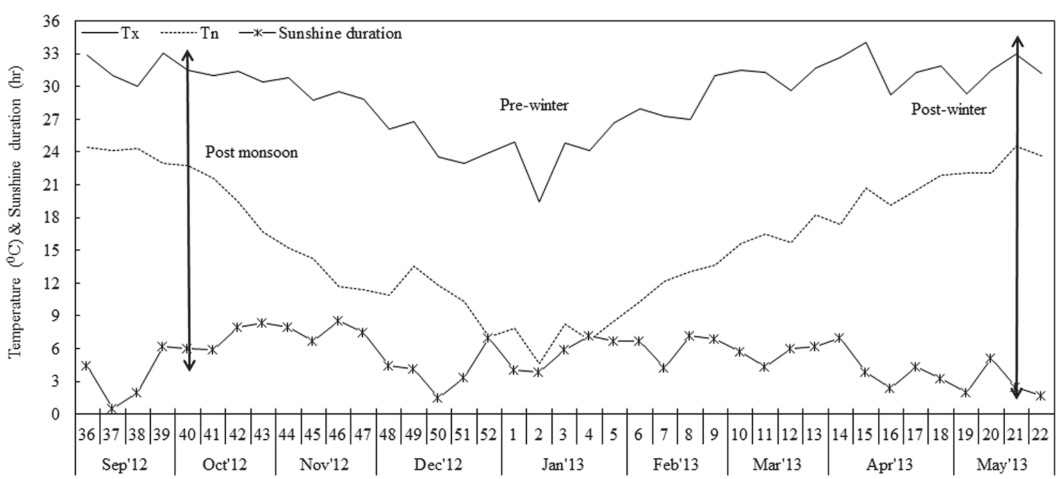

Fig. 5. Pattern of maximum and minimum temperature and sunshine duration at the chlorophyll fluorescence measurement period

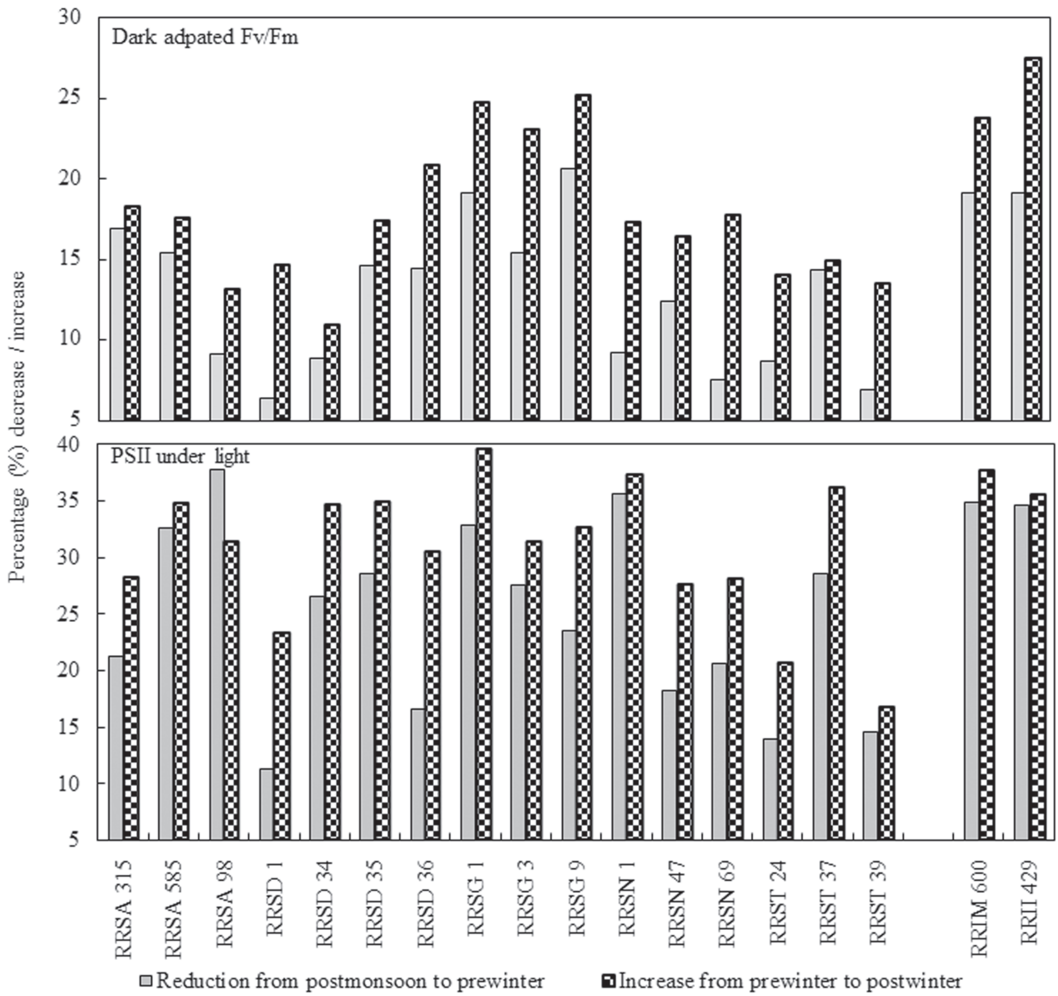

Fig. 6. Extent of depression and recovery of efficiency of excitation energy and ΦPSII upon low winter temperature stress

from post-monsoon to pre-winter was the lowest in RRSD 1 followed by RRST 39, and RRSN 69 (Fig. 6); however, the recovery percentage from the pre-winter to post-winter was the highest in RRSG 9 followed by RRSD 1 and RRSG 3. In RRSN 69 and RRSD 1, the Fv/Fm increased by 2.3 fold from pre-winter to post-winter in comparison to that of the reduction from post-monsoon to pre-winter; this was followed by RRST 39 and RRSN 1 (2.0 fold increase). However, for Φ PSII, this increase was 2.1 fold in RRSD 1 followed by RRSD 36 (1.8 fold). The increase in the value of photosynthetic efficiency (Φ PSII) was less than 1 in RRSA 98 (0.83) indicating that it was highly sensitive to the prevailing low winter temperature. Cold exposure induces physiological and biochemical changes in plants (Theocharis *et al.*, 2012; Santini *et al.*, 2013) and different degrees of resistance was developed depending on the potential of the species (Lukatkin *et al.*, 2012). Thus, clonal variation on recovery from cold stress would be an important parameter towards selecting the best acclimatized one within the population.

During pre-winter period, weekly mean temperature came down to as low as 7.1°C with sunshine duration of 6.9 hr; thus, the plants experienced low winter temperature stress and high level of irradiance. The combined effect of low temperature and high irradiance was found to be more important than the individual effect of each stress (Long *et al.*, 1994) while imparting cold injury in plant system. Inhibition in Fv/Fm and ΦPSII from warm to cold agroclimate was reported in *Hevea* clones PB 235 and RRII 105 (Alam *et al.*, 2005). Extent of decrease in Fv/Fm under dark condition due to cold stress was found to depend on cultivars (cold tolerant) in maize (Looten *et al.*, 2004). In field condition tolerance of citrus to seasonal weather changes with respect to chlorophyll fluorescence showed that during warm period Fv/Fm value was low and it was further inhibited when temperature was near to 0°C (Santini *et al.*, 2013). Multiple environmental stresses co-occur frequently under natural conditions and plants experience complex environmental stresses (Lu *et al.*, 2003) which may not be usually predictable by single-factor analyses but a combination of different environmental stress factors (Osmond *et al.*, 1986). Therefore, further study on adaptation of clones during late mature stage would be worth investigating.

CONCLUSION

The present study on evaluation of *Hevea* ortets at early stage of field establishment showed that the ortet RRSD 1, which originated from the hot climate of Daphchari, Maharashtra, was a resilient clone showing better adaptation to cold stress in terms of better survival rate in field, appreciable girth and photochemical activities. In terms of growth, the ortets collected from Guwahati were better than others. However, the clone RRSN 69 showed low apical leaf damage with stable chlorophyll content, considerable girth and better recovery rate of ΦPS II activity from pre-winter to post-winter period. On the contrary, RRSA 98 was found to be cold susceptible with high apical leaf damage, low girth and girth increment, remarkable reduction in chlorophyll content during low temperature period and slow rate of recovery of Φ PSII from pre-winter to post-winter period. These ortets with contrasting traits would be targeted for further studies to evaluate and select suitable lines for this cold prone region.

REFERENCES

Alam, B. and Jacob, J. (2002). Overproduction of photosynthetic electrons is associated with chilling injury in green leaves. *Photosynthetica,* **40:** 91-95.

Alam, B., Nair, D.B. and Jacob, J. (2005). Low temperature stress modifies the photochemical efficiency of a tropical tree species *Hevea brasiliensis:* Effects of varying concentration of CO_2 and photon flux density. *Photosynthetica,* **43**(2): 247-252.

Allen, D.J. and Ort, D.R. (2001). Impacts of chilling temperatures on photosynthesis in warm-climate plants. *Trends in Plant Science,* **6**: 36-42.

Das, G., Mondal, G.C. and Chaudhuri, D. (2013a). Adaptability of prospective mother trees of *Hevea* to the cold climate of Sub-Himalayan West Bengal and Assam. *Journal of Plantation Crops,* **41**(2): 163-71.

Das, G., Reju, M.J., Mondal, G.C., Singh, R.P., Thapliyal, A.P. and Chaudhuri, D. (2013b).

Adaptation of *Hevea brasiliensis* clones in three widely different cold prone areas of northeastern India. *Indian Journal of Plant Physiology,* **18**(3): 231-39.

Jacob, J. (2013). Chlorophyll fluorescence-based non-invasive techniques to detect and quantify climate stress on plant. *Journal of Plantation Crops,* **41**(1): 1-7.

Jacob, J., Annamalainathan, K., Alam, B., Sathik, M.M.B., Thapliyal, A.P. and Devakumar, A.S. (1999). Physiological constraints for cultivation of *Hevea brasiliensis* in certain unfavourable agroclimatic regions of India. *Indian Journal Natural Rubber Research,* **12**(1&2): 1-16.

Jiang, A. (1988). Climate and natural production of rubber (*Hevea brasiliensis*) in Xishuamg-banna, southern part of Yunan province, China. *International Journal of Biometry,* **32**: 280-282.

Jing, M., Stephane, H., Marc, V., Eric, C., Jean-Louis J., Thierry, A. and Patricia, R.D. (2010). Contrasting strategies to cope with chilling stress among clones of a tropical tree *Hevea brasiliensis*. *Tree Physiology,* **30:** 1391 -1402.

Joo, Y. and EunJu Lee, E.J. (2011). Acclimation responses of *Tamarix chinensis* seedlings related to cold stress. *Journal of Ecological Field Biology,* **34**(3): 251-257.

Kitajima, K. and Hogan, K.P. (2003). Increases of chlorophyll *a/b* ratios during acclimation of tropical woody seedlings to nitrogen limitation and high light. *Plant Cell and Environment,* **26**: 857-865.

Krause, G.H., Briantais, J.M. and Vernotte. C. (1983). Characterization of chlorophyll fluorescence quenching in chloroplasts by fluorescence spectroscopy at 77-K.1.delta-pH-dependent quenching. *Biochemical et Biophtsica Acta,* **723**: 169-175.

Krishan, B. (2013). Yield performance of elite polycross seedlings of *Hevea brasiliensis* grown in a dry sub-humid climate of India. *Rubber Science,* **26**(1): 117-122.

Long, S.P., Humphries, S. and Falkowski, P.G. (1994). Photoinhibition of photosynthesis in nature. *Annual Review of Plant Physiology and Plant Molecular Biology,* **45**: 633-662.

Looten, P., Waes, J.V. and Carlier, L. (2004). Effect of a short photoinhibition stress on photosynthesis, chlorophyll a fluorescence, and pigment contents of different maize cultivars. Can a rapid and objective stress indicator be found? *Photosynthetica,* **42**(2): 187-192.

Lu, C., Qlu, N., Wang, B. and Zhang, J. (2003). Salinity treatment shows no effects on photosystem II photochemistry, but increases the resistance of photosystem II to heat stress in halophyte *Suaeda salsa*. *Journal of Experimental Botany,* **54**(383): 851-860.

Lukatkin, A.S., Brazaityte, A., Bobinas, C. and Duchovskis, P. (2012). Chilling injury in chilling sensitive plants: a review. *Þemdirbystë Agriculture,* **99**(2): 111 124.

Mai, J., Herbette, S., Vandame, M., Cavaloc, E., Julien, J.-L., Améglio, T., and Roeckel-Drevet, P. (2010). Contrasting strategies to cope with chilling stress among clones of a tropical tree, *Hevea brasiliensis*. *Tree Physiology,* **30**: 1391–1402.

Mai, J., Herbette, S., Vandame, M., Kositsup, B., Kasemsap, P., Cavaloc, E., Julien, J.-L., Améglio, T. and Roeckel-Drevet, P. (2009). Effect of chilling on photosynthesis and antioxidant enzymes in *Hevea brasiliensis* Muell. Arg. *Trees,* **23**: 863–874.

Nair, D.B. and Jocob, J. (2011). A simple method of large scale estimation of leaf chlorophyll content in *Hevea brasiliensis* using chlorophyll meter. *North Bengal University Journal of Plant Science,* **5**(1): 47-49.

Osmond, C.B., Austin, M.P., Berry, J.A., Billings, W.D., Boyer, J.S., Dacey W.J.H., Nobel, P.S., Smith, S.D. and Winner, W.E. (1986). Stress physiology and the distribution of plants. *BioScience,* **37**: 38-48.

Raj, S., Das, G., Pothen, J. and Dey, S.K. (2005). Relationship between latex yield of *Hevea brasiliensis* and antecedent environmental parameters. *International Journal of Biometeorology,* **49**:189–196.

Ray, D. , Dey, S.K. and Das, G. (2004). Significance of the leaf area ratio in *Hevea brasiliensis* under high irradiance and low temperature stress. *Photosynthetica,* **42**(1): 93-97.

Renaut, J, Hoffmann, L and Hausman, J.F. (2005). Biochemical and physiological mechanisms related to cold acclimation and enhanced freezing tolerance in poplar plantlets. *Physiologia Plantarum,* **125**: 82-94.

Rinne, P.L.H., Kaikuranta, P.M. and Schoot, C.van der. (2001). The shoot apical meristem restores its symplasmic organization during chilling-induced release from dormancy. *The Plant Journal,* **26**(3): 249-264

Santini, J., Giannettini, J., Pailly, O., Herbette, S., Ollitrault, P., Berti, L. and Luro, F. (2013). Comparison of photosynthesis and antioxidant performance of several *Citrus* and *Fortunella* species (Rutaceae) under natural chilling condition. *Trees – Structure and Function,* **13**: DOI: 10.1007/s00468-012-0769-5

Schreiber, U., Bilger, W., Hormann, H. and Neubauer, C. (1998). Chlorophyll fluorescence as a diagnostic tool: basics and some aspects of practical relevance. In: *Photosynthesis: A Comprehensive Treatise*. (Ed. A.S. Raghavendra). Cambridge University Press, Cambridge. pp.320-336.

Sevillano, L., Sanchez-Ballesta, M.T., Romojaro, F. and Flores, F.B. (2009). Physiological, hormonal and molecular mechanisms regulating chilling injury in horticultural species. Post-harvest technologies applied to reduce its impact. *Journal of Science, Food and Agriculture,* **89**: 555–573.

Theocharis, A., Clement, C. and Barka, E.A. (2012). Physiological and molecular changes in plants grown at low temperature. *Planta,* **235**(6): 1091-1105.