GAS EXCHANGE CHARACTERISTICS AND CHLOROPHYLL-*a* FLUORESCENCE KINETICS OF TWO MORPHOTYPES OF *CENTELLA ASIATICA* UNDER OPEN AND SHADED CONDITIONS

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SUMMARY

The differences in leaf Ψw, pigment levels and photosynthetic activity of two contrasting leaf morphotypes (PT1 and PT2) of Mandukaparni (*Centella asiatica* L.) grown in open (sun) and under artificial green shade nets of 25% and 67% shade levels were investigated. Open and shade plants of PT1 and PT2 showed similar diurnal fluctuations in Ψw. The Chl *a*, Chl *b*, carotenoid content and Chl: carotenoid ratio in plants under 67% shade were 1.53, 1.63, 1.10 and 0.87 times of open plants in PT1 whereas PT2 had 1.86, 1.96, 1.37 and 0.69 times the respective value under shade. PT1 recorded much lower net photosynthetic rate (P_n) compared to PT2 and the peak P_n in PT1 was 66.7% and 57.1% of PT2 in open and 25% shade, respectively. Fluctuation in stomatal conductance (g_s) during the day was more acute in PT2 compared to PT1 and g_s closely coupled with P_n at different light intensity in PT2 compared to PT1. Both the plant types recorded higher transpiration (*E*) in 25% shade. Chlorophyll fluorescence values were significantly altered under low light levels although primary photochemistry was least affected by high light. Efficiency of photosystem II (ΦPS II) increased by 4.3 and 3.6 times of open under 67% shade in PT1 and PT2, respectively. The photochemical quenching (q*P*) increased substantially under 67% shade with 48% and 42% increase compared to open in PT1 and PT2 respectively, whereas non photochemical quenching (q*N*) increased by 22% and 30% in open compared to 25% and 67% shade. Increase in shade resulted in reduction in electron transport rate (ETR) value in both plant types. Leaf size increased 3.38 and 1.54 times in PT2 and PT1 under 67% shade. Specific leaf area (SLA) also increased by 1.31 and 1.42 times under 25% and 67% shade respectively, in PT1. Results revealed that PT2 was less sensitive to high light and exhibited higher plasticity to varying light intensity.

***Key words: Centella asiatica*, chlorophyll fluorescence, gas exchange, leaf growth, light intensity, shade**

INTRODUCTION

Centella asiatica (L.) Urb. (syn.: *Hydrocotyle asiatica* L.) of the Apiaceae (Umbelliferae) family is a small herbaceous creeping perennial plant, native to India, China, Indonesia, Australia, the South Pacific, Madagascar, and southern and middle Africa. It is commonly known as, Mandukaparni, Gotu Kota and Indian Pennywort. In Ayurvedic system of medicine, it is recommended for chronic diseases and as a "brain tonic" for various mental disorders. It is listed officially in the Chinese Pharmacopoeia and used as diuretic and

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antipyretic, and in the treatment of icterus, heatstroke, diarrhea, ulcerations, eczema, and traumatic diseases (Tang and Eisenbrand 1992, Kartnig 1998). Recently, clinical efficacy of *C. asiatica* to attenuate the age-related decline in cognitive function in healthy middle age and elderly adults has been experimentally proved (Dev *et al*. 2009). It contains various bioactive principles such as asiaticoside, centelloside, madecassoside, brahmoside, brahminoside, thankuniside, sceffoleoside, centellose, and asiatic, brahmic, centellic and madecasic acids (Aziz *et al.* 2007). Depending on the origin of the plant material, these saponins can account for 1 to 8% of the constituents (Brinkhaus *et al.* 2000).

Light intensity at extreme levels becomes limiting factor for photosynthetic carbon gain. Photoinhibition occurs at full sunlight, however, at very low light levels, photosynthesis is limited by available photon flux density (PFD). Thus, shade tolerant plants are adapted to survive under conditions that plants adapted to sunlight would not be able to survive. Normally, when plants occupy a range of microenvironments from sunlight to shaded, they exhibit a highly plastic series of compromises that function to minimize the potential for photoinhibition when exposed to high PFD but maximize light capture when the plant is in the shade (Pearcy and Sims 1994). This plasticity, which at the leaf-physiological level takes the form of photosynthetic sun/shade acclimation and photoprotection *via* energy dissipation, differs with plant species and within species as well (Pearcy and Sims 1994, Demmig-Adams and Adams 1992). According to Rajcan and Swanton (2001) the quality and quantity of intercepted light is correlated with plant morphology. Plant morpho-types which differ in leaf size, leaf characteristics and triterpenoid saponin content have been reported in *C. asiatica* (Jacinda *et al.* 2008, Zainol *et al.* 2008). In our study, we examined the physiological acclimation of *C. asiatica* morphotypes with contrasting leaf characters to the light environments by studying diurnal changes in leaf water relationships, photosynthetic rate (P_n) and parameters of Chl *a* fluorescence.

MATERIALS AND METHODS

Plant growth conditions: The experiment was conducted under field conditions during two consecutive years 2008-09 and 2009-10 at the research farm of Directorate of Medicinal and Aromatic Plants Research (22°35'36'' N 72°56'08'' E). *Centella asiatica* plants with contracting leaf types, i.e. PT1 with small leaves (6-7 cm^2) and PT2 (var. Vallabh-Medha, developed at DMAPR, Anand) with large leaves (15-20 cm^2) were grown under irrigated conditions under open and artificial green shade nets of 25% and 67% shade levels. The design was completely randomized with four replications. Stem cuttings of both small leaf type and big leaf type were planted at 30x60 cm spacing in leveled plots of 4x4 m.

Leaf water potential measurement: Leaf water potential (Ψw) was measured using pressure chamber (Soil Moisture Corporation, Santa Barbara, U.SA). Ψw measurements were taken between 6.00 and 18.00 hrs on fully expanded leaves. The end of leaf petiole was cut by sharp blade and kept inside the chamber with petiole exposed. The chamber was then sealed and pressure applied slowly. The balance pressure required to force the sap to the cut surface was taken as measure of Ψw.

Measurement of gas exchange, chl fluorescence and photosynthetic pigments: The parameters of gas exchange were measured on middle, fully expanded leaves of plants between 06:00 and 18:00 during bright sunlight on a clear, cloudless day (average T_{air} was 27°C, average PPFD was approximately 1800 μmol m^{-2} s^{-1}, and average air relative humidity 40%, content of soil water was lower than 30 %. The parameters included P_n, transpiration rate (E), stomatal conductance (g_s) and intercellular CO_2 concentration (C_i), that were measured using a portable photosynthesis system (*LI-6400*, *LI-COR*, Lincoln, USA). Two leaves were analyzed from each plant one time, and four replications were taken per treatment.

Chl *a* fluorescence parameters were monitored under field conditions using a portable pulse amplitude modulation fluorometer (*LI-6400-40, LI-COR*, Lincoln, USA). Minimal and maximal fluorescence of a dark adapted leaf at 11.00 hrs after a 60 min dark adaptation was observed to calculate maximum photochemical efficiency (F_v/F_m). The photosynthetic photon flux density (PPFD) was kept coinciding with natural light of 1800 μmol m^{-2} s^{-1}. Efficiency of energy harvesting in the light

(F_v'/F_m'), values of the effective quantum yield of PSII (ÖPSII), photochemical quenching (qP), non-photochemical quenching (qN), and electron transport rate values (ETR) were measured. The results represented are means ± SD of the measurements made on 8 different leaves (two leaves per plant). Chlorophyll and carotenoid content of the fully expanded mature leaf (0.2 g per replication) was determined after extraction with 80% acetone (Lichtenthaler and Buschmann 2001).

Statistical analysis: Data were subjected to analysis of variance (ANOVA) following RCBD model using MSTAT-C software version 1.4 (Crop and Soil Science Division, Michigan State University, USA). Means were compared with least significant difference (P=0.05). Two repetitions of the experiments were analyzed separately and an average data from one experiment are presented here as results from both the experiments followed similar trend.

RESULTS

Photosynthetic pigments: Photosynthetic pigments content was affected significantly (P < 0.05) by the different light intensity treatments (Fig. 1). The open light and 25% shade treatment resulted in significant reductions in Chl *a*, Chl *b* and total chlorophyll content in both PT1 and PT2. The highest Chl *a*, Chl *b* and total chlorophyll content and the lowest Chl *a*:*b* ratio were observed in the 67% shade treated plants. Significantly higher chl *a*, chl *b* content were observed in PT2 compared to PT1 under 25% and 67% shade. Chl *a*, Chl *b*, carotenoid content and Chl: carotenoid ratios in 67% shade treatment plants were 1.53, 1.63, 1.10 and 0.87 times of open plants of PT1 whereas PT2 had 1.86, 1.96, 1.37 and 0.69 of open. Ratio of chl: carotenoid decreased under shade in both the plant types. The reduction was 15.33% and 44.51% in 25% and 67% shade respectively, compared to open plants of PT2.

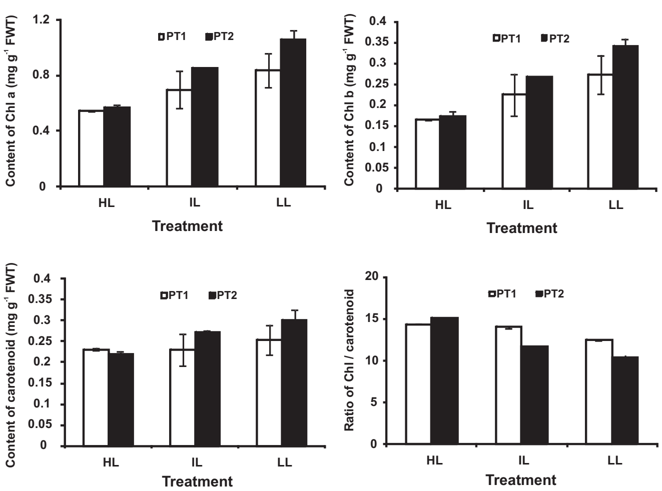

Fig. 1. Comparison of chlorophyll (Chl) content and carotenoid content in leaves of *C. asiatica* grown under full sunlight, 25% and 67% shade. Values are means ± S.D. (HL- open, IL– 30% shade, LL– 67% shade)

Diurnal changes in leaf water potential (Ψw): Diurnal changes in leaf water potential (Ψw) showed similar pattern in both PT1 and PT2 in all light levels studied. At all light intensities, PT1 showed comparatively higher water status than PT2 during the entire day. In both the plant types, Ψw was at -0.2 MPa at 6.00 hrs and steadily declined afterwards to reach -1.2 MPa in both open and 25% shade (Fig. 2). At open light condition the decline in Ψw was marginal after 14.00 hrs, whereas, 25% and 67% shade did not show any change up to the 18.00 hrs. The leaves did not show any symptom of wilting in the afternoon hours, even though there was a slight upward curling of leaves of both the plant types under open conditions. The recovery in Ψw was not observed even at 18.00 hrs in plants under the different light intensities studied.

Gas exchange parameters: Regardless of shading treatment, PT2 had higher P_n values compared to PT1. The P_n showed midday depression in both the plant types across the light regimes. Net P_n decreased with increasing shade levels. P_n in PT2 increased very quickly early in the morning and then decreased sharply, reaching a minimum at about 12:00, whereas PT1 had the peak at 14.00. PT1 recorded much lower P_n compared to PT2 and the peak P_n in PT1 was 66.7% and 57.1% of PT2 in open and 25% shade, respectively. Reduction in P_n was negligible at 67% shade in PT1 whereas, PT2 had only 58.84% compared to open at 12:00. The day time fluctuations in P_n were less pronounced in PT1 and consistently lower than PT2 at all the light regimes. At 12:00 hrs under 67% shade, P_n recorded 58.84% and 81.80% of PT2 and PT1, respectively. Substantial reduction in P_n was noticed in PT2 whereas PT1 showed only negligible decline even at higher shade.

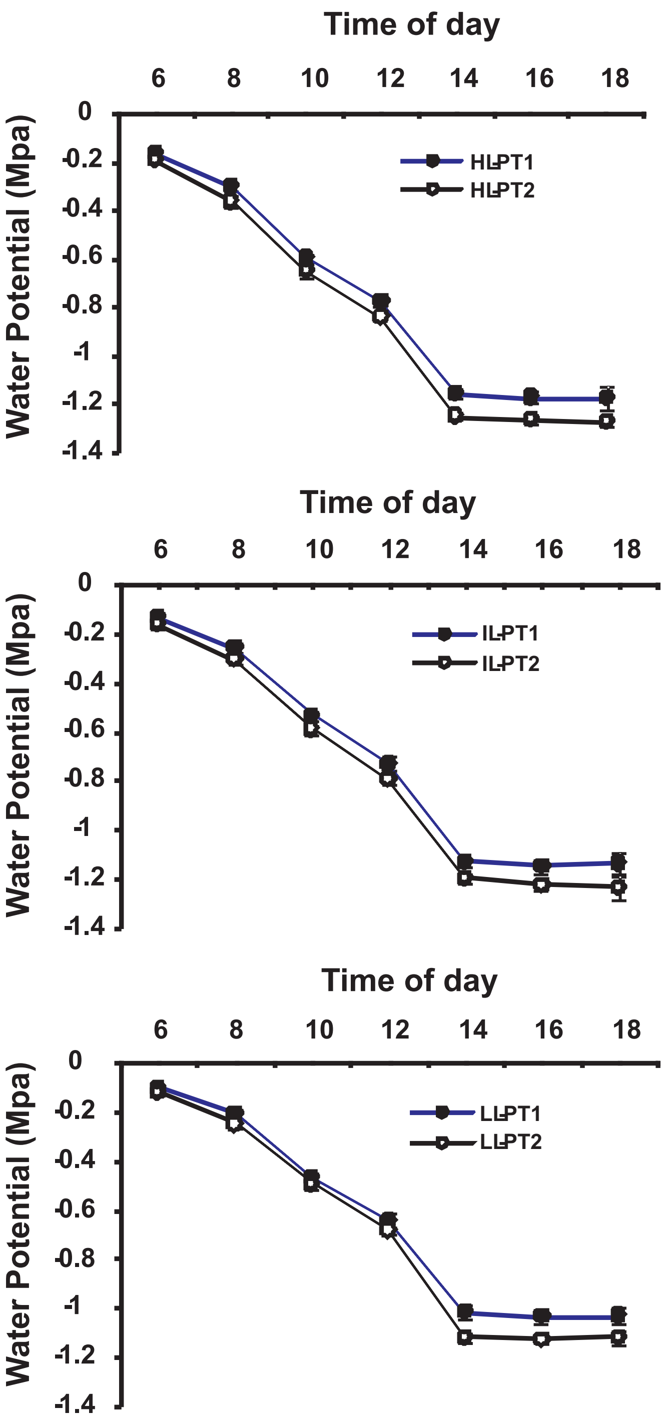

Fig. 2. Diurnal course of leaf water potential (Ψw) of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

The g_s followed similar trend as P_n in both the plant types. The data on g_s showed slight midday depression in both the morphotypes for all the light levels studied (Fig. 3). Fluctuation in g_s was more prominent in PT2 compared to PT1. The P_n and g_s closely coupled to light intensity in PT2 compared to PT1. Peak g_s recorded

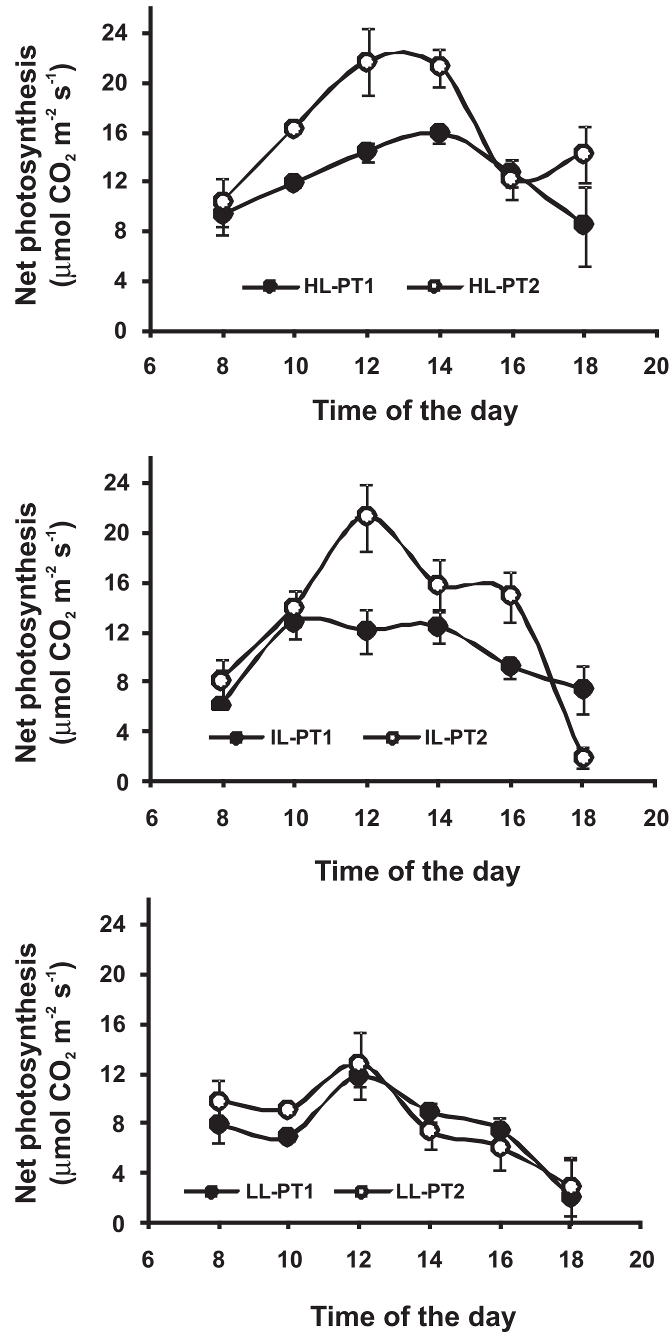

Fig. 3. Diurnal course of P_n of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

significantly higher in PT2 compared to PT1 at 12.00. Transpiration (*E*) showed marked increase in the morning hours with a peak rate at 12:00 in both PT1 and PT2 in all the light levels. However, a second peak was noted at 16.00 hrs. under shade in PT2. Plants under 25% shade recorded higher *E* compared to the other light levels. Plants grown under higher shade levels had smaller daily transpiration total than open light because of the higher g_s. Leaf measurements of g_s of the high light plants showed that g_s decreased substantially at low PFD. The g_s and *E* showed decreasing trend from noon coinciding with sharp decline in leaf water potential.

Chl a fluorescence parameters: The F_v/F_m (maximum quantum efficiency of PS2) did not show significant changes under open light and shade conditions implying that high light intensity did not alter the primary photochemistry. However, significant increase in ΦPS2 was recorded under shade in both PT1 and PT2 with 2 fold increase at 25% shade (Table 1). At 67% shade, it increased by 4.3 and 3.6 times of open in PT1 and PT2, respectively. Photochemical quenching (q*P*) increased substantially at 67% shade with values 48% and 42% more compared to open in PT1 and PT2, respectively. Among the light levels, q*N* was highest in open and increased proportionately with increasing light intensity. PT2 recorded higher q*N* compared to PT1 at 25% and 67% shade with 22% and 30% increase over open light. ΦCO_2 (quantum yield of CO_2 assimilation) increased several fold at 67% shade compared to open in both PT1 and PT2 (Table 1). It was 4.7 and 2.8 times of open in PT1 and PT2 at 12.00 hrs when the Pn reached maximum values. Highest values of F_v'/F_m' (PSII quantum yield) were recorded at early morning in open and 25% shade. It progressively decreased during the day to recover only in the late evening after 16.00 hrs. The F_v'/F_m' values were consistently higher under shade compared to open. The values of F_v'/F_m' were decreased by 53% and 47% at 12.00 hrs in both PT1 and PT2 in open compared to 67% shade. However, the fall in F_v'/F_m' in plants under 25% shade at midday was less and they maintained 60% and 67% of 67% shaded plants in PT1 and PT2 at 12.00 hrs. Significant alteration in ETR was observed under shade in both PT1 and PT2. ETR

Table 1. Changes in leaf characteristics and Chl *a* fluorescence parameters of *C. asiatica* plant grown under different light intensity.

Parameter	PT1			PT2			C.D (A) P=(0.05)	C.D (B) P=(0.05)	C.D (A*B) P=(0.05)
	Open	25% shade	67% shade	Open	25% shade	67% shade			
LA (cm^2)	6.20	8.25	9.54	17.14	33.12	57.92	0.295	0.241	0.042
Dry weight of leaf (g)	0.036	0.037	0.039	0.099	0.184	0.248	0.005	0.004	0.001
Specific leaf area (cm^2g^{-1})	172.27	225.10	245.18	180.10	233.31	172.17	6.145	5.02	0.87
Specific leaf weight (mg cm^{-2})	5.805	4.442	4.079	5.553	4.286	5.808	0.1382	0.1	0.02
F_v/F_m	0.781	0.791	0.777	0.808	0.804	0.763	0.009	0.007	0.013
PhiPS2	0.096	0.198	0.414	0.224	0.423	0.116	0.005	0.004	0.001
q_p	0.505	0.544	0.748	0.584	0.763	0.535	0.009	0.007	0.001
q_n	0.950	0.887	0.729	0.870	0.684	0.938	0.005	0.004	0.001
PhiCO2	0.00925	0.013	0.04375	0.0135	0.021	0.03825	0.001	0.0004	0.001

CD (A) = Plant type , CD (B) = Light level , CD (A*B) = Plant type x light level

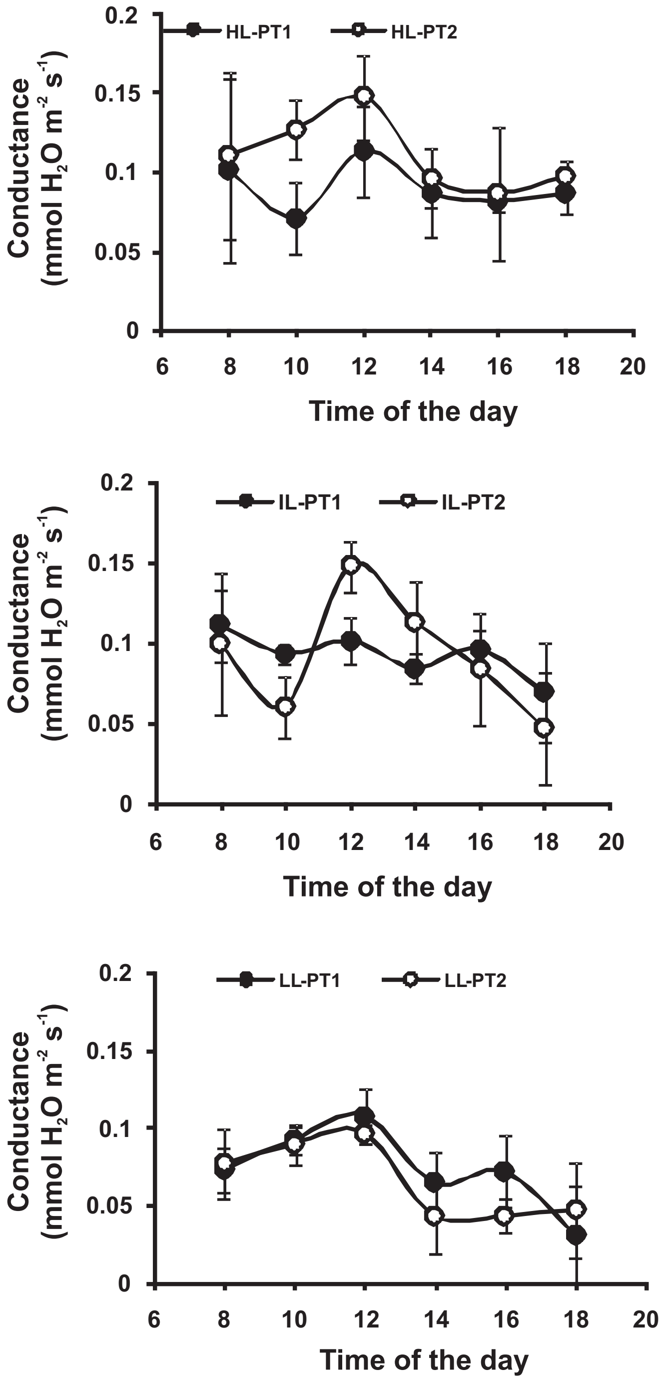

Fig. 4. Diurnal course of leaf conductance (g_s) of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

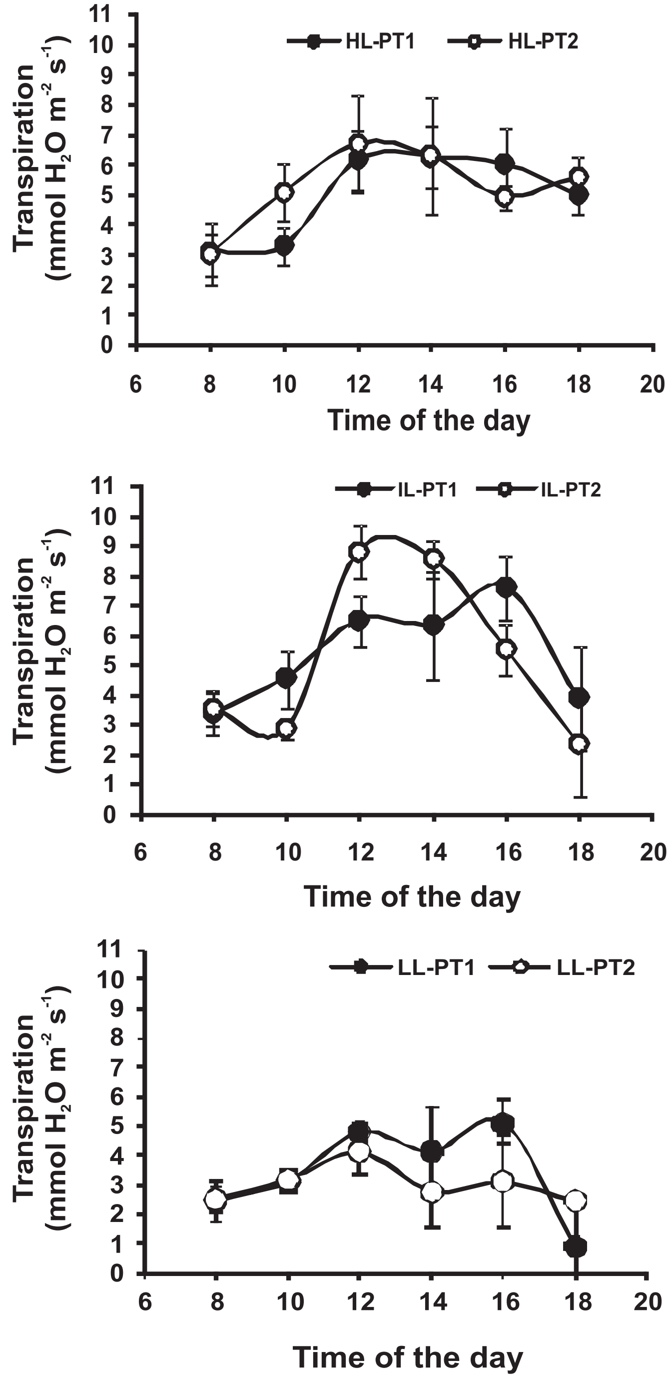

Fig. 5. Diurnal course of transpiration (E) of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

values were relatively stable all through the day in plants grown under open. Increase in shade resulted in reduction in ETR value and 67% shade maintained ETR values of 79% and 70% of open at 12:00 hrs. in PT1 and PT2. ETR values in 25% shade were also lower in early morning and evening hours when light intensity was low.

Leaf characteristics: Significant changes in leaf growth characteristics of both the plant types were observed in open and shade. Leaf size increased in both PT1 and PT2 under shade. Leaf size increased by 1.93 and 3.38 times of open in 25% and 67% shade in PT2, respectively, whereas the increase was 1.33 and 1.54 times in PT1 for the 25% and 67% shade plants (Table 1). Petiole length also increased with shade and the increase was higher for 67% shade plants of PT1 (1.94 times) and PT2 (2.12 times) compared to open. Similarly, dry weight of individual leaf increased substantially under shade with 1.84 and 2.49 times of open light whereas, PT1 did not show appreciable increase in leaf dry weight. At 25% and 67% shade, PT1 recorded higher specific leaf area (SLA) which was 1.31 and 1.42 times of open whereas, PT2 recorded 35% higher SLA at 67% shade compared to open. Specific leaf weight (SLW) showed significant reduction in shade in both the plant types. Lowest SLW was recorded in 67% shade plants of PT1 which was 30% lower compared to open, whereas it reduced by 27% in 67% shade plants of PT2.

DISCUSSION

Light is one of the most important determinants of growth and productivity of field crops and cumulative intercepted radiation is often linked to plant biomass production (Monteith, 1994). Genotypes differ in their response to incident light through physiological functions as well as growth and development. Results showed that leaf water relations and leaf gas exchange and other physiological parameters were altered to varying degrees in both the plant types of *C. asiatica* grown under light and shade. Leaf water potential (Ψw) at midday reached -1.2 MPa in plants of both PT1 and PT2 under shade and open, typical of irrigated plants under field conditions. PT1 maintained comparatively higher Ψw throughout the day probably due to smaller leaf size and lower transpiration (*E*). Photosynthetic metabolism is inhibited by decrease in Ψw and with increasing stress intensity, stomatal limitations cause further decline in net CO_2 assimilation rate (Lawlor and Cornic 2002). It is known that due to excess atmosphere water demand water stress is experienced even by irrigated plants at midday (Chaves 1991) and P_n is down regulated. The fall in leaf water potential was not significantly different from open in shade plants even though the transpiration rate was higher in open and 25% shade. The g_s followed the diurnal time course of PPFD during the whole morning. During the afternoon, however there was a consistent decrease in P_n that matched a decrease in g_s. Interestingly higher leaf water potential recorded in PT1 did not translate into higher g_s compared to PT2. Relatively higher g_s and E with high Ψw in PT1 may be attributed to the lower reduction in P_n under shade compared to PT2. Transpiration rates per unit leaf area of the open grown plants were approximately twice those of shade plants. The reduced SLA of the sun plants compensated for their higher transpiration rates per unit leaf area. However, nearly three fold increase in leaf size resulted in higher transpiration loss even under shade.

Jason *et al*. (2004) described the reduction in chl *b* content in excess radiation as an indication of chlorophyll destruction. Severe reduction in Chl *a* and Chl *b* of PT2 suggests that it is well adapted to shade rather than high light intensities. Although, chlorophyll content was markedly reduced under open and 25% shade, higher P_n was noticed in these treatments compared to low light condition. Substantial increase in chlorophylls and carotenoid content in 67% shade indicate the ability of plants to optimize the light harvesting at low light conditions (Lei *et al.* 1996). Leaf chlorophyll content also is one of the most important factors in determining the photosynthesis rate (Mao *et al.* 2007) and dry matter production (Ghosh *et al.* 2004).

Leaves grew larger when grown under 67% shade (Table 1) in both the plant types. Many shade loving plants tend to increase the leaf size under low light conditions to maximize the light harvesting to help greatly decreased the photosynthetic capacity of the leaves (Campbell and Miller 2002). The increase in leaf size was greater at 67% in PT2 (57.92 cm^2) with concurrent increase in dry weight (0.248 g) indicated that it well adapted to low light intensities compared to PT1. Marked increase in ΦCO_2 under low light intensity (67% shade) in both PT1 and PT2 showed that the adaptation of *C. asiatica* to low light.

Quantum yields of high and low PPFD grown *A. macrorrhiza* plants had also shown similar results (Chow *et al.* 1988, Sims and Pearcy 1989), as in our

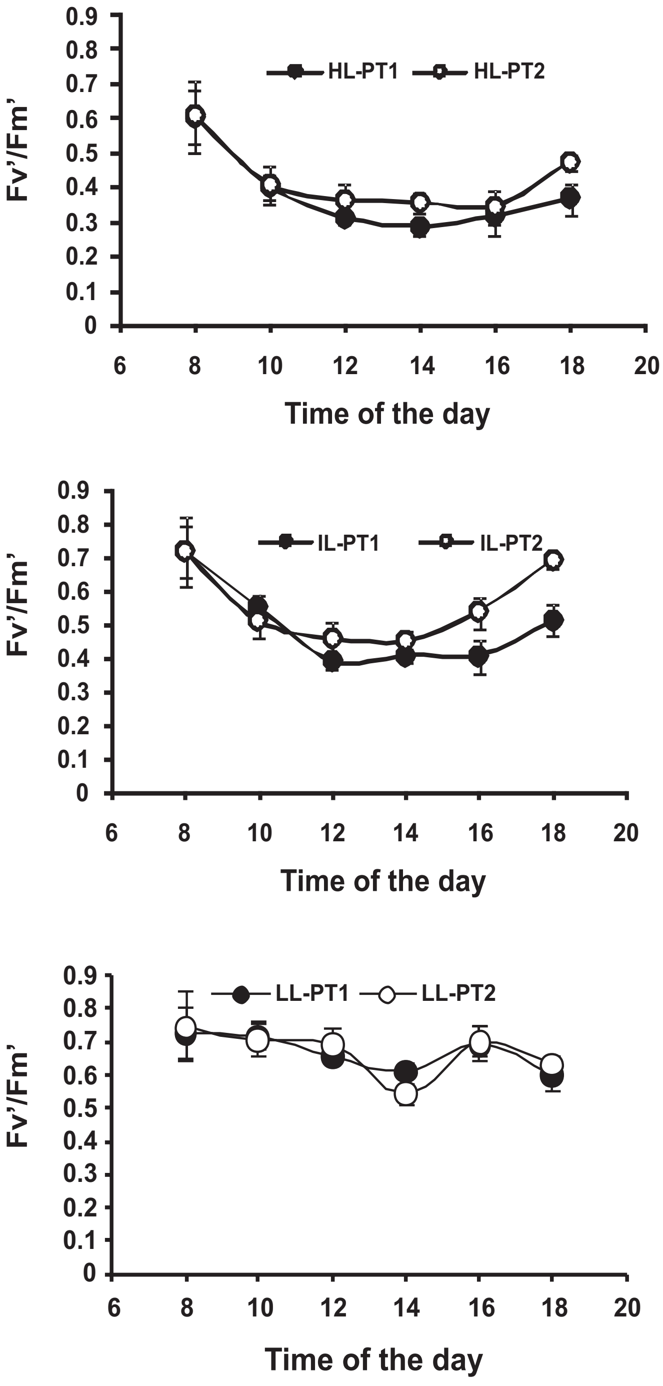

Fig 6: Changes in Fv'/Fm' of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

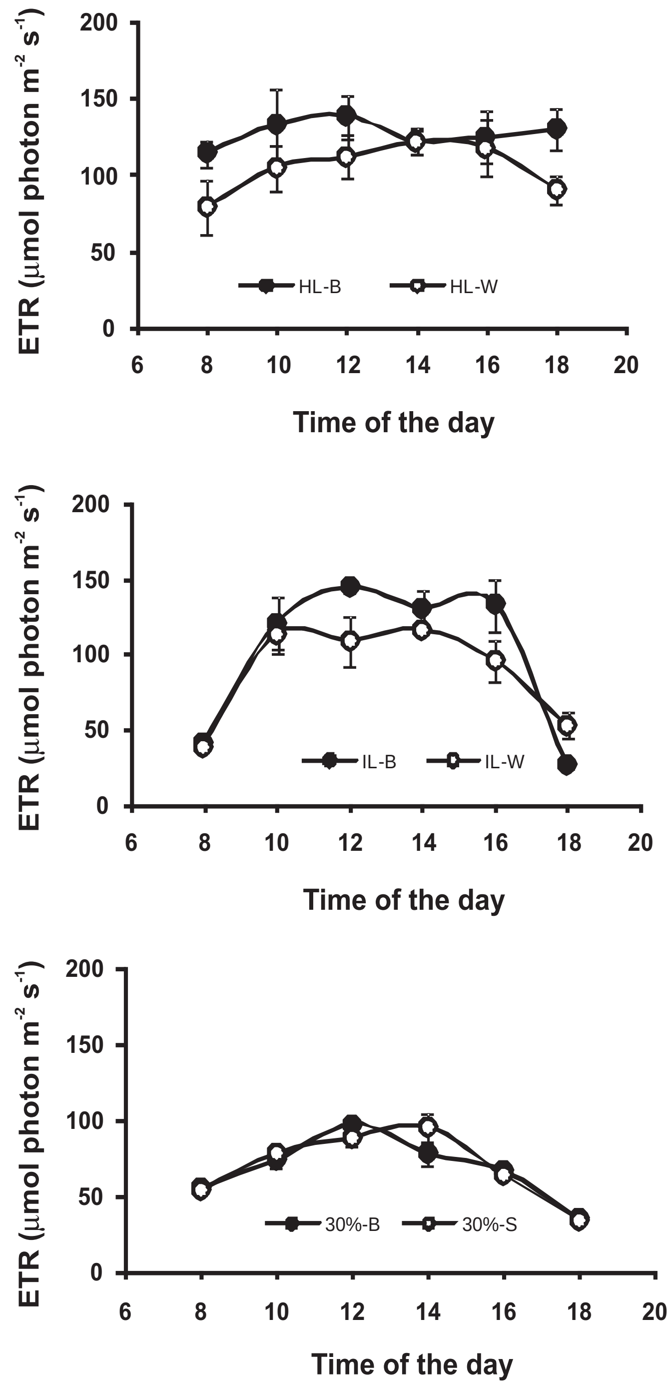

Fig. 7. Changes in ETR (μmol photon m^{-2} s^{-1}) of *C. asiatica* plant types grown at different light intensities (Values are means ± S.D, HL- open, IL – 30% shade, LL – 67% shade)

study at whole plant level. In our study, high and low light intensity does not alter the primary photochemistry of *C. asiatica* as evidenced by minimal changes in F_v/ F_m at high light intensity. Thus, superior light capture capacity afforded by the higher SLA under shade/low light provides a substantial advantage. At high light intensity, the fluorescence parameters like F_v'/F_m' (PSII maximum efficiency) and ETR were higher in PT2 compared to PT1 which suggested that PT2 was less sensitive to high light conditions. As high photochemical quenching translate into high ETR and quantum yield observed in our experiment in PT2 under high light, the damage to photosystems is avoided by high q*N* by dissipating the excess energy as heat in both plant types and prevents the PS2 from being hampered by high irradiance. The efficiency of PSII is decreased leading to reduced rate of photosynthesis due to the oxidative damages caused by high light (Niyogi 1999). Significant decrease in photosynthetic pigments corroborate the decrease in P_n at midday due to the decreased F_v'/F_m', qP and increased qN in leaves of *C. asiatica* under open and 25% shade. At high daily PPFD, however, higher photosynthetic capacity per unit leaf area achieved by open plants with decreased SLA and the advantage of carbon gain per unit biomass is limited by the reduced leaf area. Per unit carbon assimilation in terms of P_n was higher under high light however, water relations, leaf growth and biomass accumulation was enhanced under shade. PT2 (var. Vallabh-Medha) was less sensitive to high light stress and exhibited higher plasticity to varying light intensity as evidenced by superior leaf growth and enhanced florescence parameters and leaf biomass production capacity.

REFERENCES

Aziz, Z.A., Davey, M.R., Power, J.B., Anthony, P., Smith, R.M. and Lowe, K.C. (2007). Production of asiaticoside and madecassoside in *Centella asiatica in vitro* and *in vivo*. *Biol. Plantarum*. **51:** 34-42.

Brinkhaus, B., Linder, M., Schuppan, D. and Hahn, E.G. (2000). Chemical, pharmacological and clinical profile of the East African medicinal plant *Centella asiatica*. *Phytomed.* **7:** 427-448.

Campbell, S.J. and Miller, C.J. (2002). Shoot and abundance characteristics of the seagrass *Heterozostera tasmanica* in Westernport estuary (south-eastern Australia). *Aqua. Bot.* **73:** 33-46.

Chaves, M.M. (1991). Effects of water deficits on carbon assimilation. *J. Exp. Bot.* **42:** 1-16.

Chow, W.S., Luping, Q., Goodchild, D.J. and Anderson, J.M. (1988). Photosynthetic acclimation of *Alocasia macrorrhiza* (L.) G Don to growth irradiance: structure, function and composition of chloroplasts. *Aust. J. Plant Physiol.* **15:** 107-122.

Demmig-Adams, B. and Adams, W. (1992). III: Photoprotection and other responses of plants to high light stress. – *Ann. Rev. Plant Physiol. Plant Mol. Biol.* **43:** 599-626.

Dev, R.D.O., Mohamed, S., Hambali, Z. and Samah, B.A. (2009). Comparison on cognitive effects of *Centella Asiatica* in healthy middle age female and male volunteers. *Europ. J. Sci. Res.* **31:** 553-565.

Ghosh, P.K., Ajay Bandyopadhyay, K.K., Manna, M.C., Mandal, K.G., Misra, A.K. and Hati, K.M. (2004). Comparative effectiveness of cattle manure, poultry manure, phosphocompost and fertilizer-NPK on three cropping systems in vertisols of semi-arid tropics. II. Dry matter yield, nodulation, chlorophyll content and enzyme activity. *Bioresou. Tech.* **95:** 85-93.

Jacinda, T.J., Meyer, R., Dubery, I.A. (2008). Characterisation of two phenotypes of *Centella asiatica* in Southern Africa through the composition of four triterpenoids in callus, cell suspensions and leaves. *Plant Cell Tissue and Organ Culture*. **94:** 91-99.

Jason, J.G., Thomas, G.R. and Pharr, D.M. (2004). Photosynthesis, chlorophyll fluorescence, and carbohydrate content of illicium taxa grown under varied irradiance. *J. Amer. Soc. Hort. Sci.* **129:** 46-53.

Kartnig, T. (1998). Herbs, Spices and Medicinal Plants, vol. 3, L.E. Cracker, J.E. Simon (Eds.), Oryx Press, Arizona, USA, pp. 145-173.

Lawlor D.W. and Cornic, G. (2002). Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. *Plant, Cell Environ.* **25:** 275-294.

Lei, T.T., Tabuchi, R., Kitao, M. and Koike, T. (1996). The functional relationship between chlorophyll content, leaf reflectance, and light capturing efficiency of Japanese forest species under natural shade and open light regimes. *Physiol. Planta*. **96:** 411-418.

Lichtenthaler, H.K. and Buschmann, C. (2001). Chlorophyll and caroteinoids: Measurement and characterization by UV-

Vis Spectroscopy. *Curr. Proto. Food Analytic. Chem.* **F4.3**: 1-8.

Mao, L.Z., Lu, H.F., Wang, Q. and Cai, M.M. (2007). Comparative photosynthesis characteristics of *Calycanthus chinensis* and *Chimonanthus praecox*. *Photosynthetica*. **45:** 601-605.

Monteith, J.L. (1994). Validity of the correlation between intercepted radiation and biomass. *Agric. For. Meteorol*. **68:** 213-220.

Niyogi, K. (1999). Photoprotection revisited: Genetic and molecular approaches. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* **50:** 333-359.

Pearcy, R.W. and Sims, D.A. (1994). Photosynthetic acclimation to changing light environments: scaling from the leaf to the whole plant. In: Caldwell, M.M., Pearcy R.W. (eds.) Exploitation of environmental heterogeneity by plants: ecophysiological processes above and below ground. Academic Press, San Diego, pp 145-174.

Rajcan, I. and Swanton, C.J. (2001). Understanding maize–weed competition: resource competition, light quality and the whole plant. *Field Crop Res.* **71:** 139-150.

Sims, D.A. and Pearcy, R.W. (1989). Photosynthetic characteristics of a tropical forest understory herb, *Alocasia macrorrhiza*, and a related crop species, *Colocasia esculenta* grown in contrasting light environments. *Oecologia.* **79:** 53-59.

Tang, W. and Eisenbrand, G. (1992). Chinese drugs of plant origin, Springer-Verlag, Berlin. pp. 273-276.

Zainol, N.A., Voo, S.C., Sarmidi, M.R. and Aziz, R.A. (2008). Profiling of *Centella asiatica* (L.) urban extract. *The Malay. J. Analytic. Sci.* **12:** 322-327.