Varied Mechanisms of Gas Exchange Parameters in Response to Altered Moisture Levels in Drought-Tolerant Cluster Bean [Cyamopsis Tetragonolaba (L.) Taub.] Genotypes

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Abstract: An effort was made at Regional Agricultural Research Station (RARS), Rekulakunta to study the physiological attitudes of cluster bean (*Cyamopsis tetragonoloba*) against different water regimes such as 100%, 80%, 60%, 40%. Six cluster bean genotypes were grown in pots in a complete random block design (CRBD) and were regularly irrigated. Plants were subjected to drought treatment at 39 days after sowing (DAS) for 8 days. On the 46th day of sowing photosynthetic rate (P_N), stomatal conductance (g_S), transpiration (E), internal CO_2 (C_i) were measured. Intrinsic water use efficiency (WUE_i), relative water content (RWC), SPAD chlorophyll meter reading (SCMR) and specific leaf area (SLA) were determined using standard protocols. The maximum P_N, g_S, E, and C_i were recorded at 60% water level coupled with RWC, SCMR, and SLA. With the increased drought, genotypes showed reduced P_N, g_S, E, and C_i except in varieties RGC-1025 and RGC-936. WUE_i showed a negative correlation with P_N, g_S, E, and C_i indicating the genotypical efficiency in copping water stress.

Keywords: Assimilation; Chlorophyll Density; Photosynthesis; IRGA; Varieties.

1. INTRODUCTION

Plants are constantly exposed during their life cycle to adverse environmental conditions that negatively affect growth, development, or productivity. Cluster bean (*Cyamopsis tetragonoloba* L.) is generally considered as a drought tolerant crop. Some studies have revealed that water stress experienced during critical growth stages can lead to significant reduction of crop yields [1,2]. To get the enhanced yields the availability of moisture during flowering and pod filling stages more important [3]. Water stress during both flowering and pod-filling stages may reduce the crop yields [4].

A close relation between SCMR and chlorophyll density reported earlier [5-9]. Chlorophyll density and SCMR association might be influenced by the factors such as time of observation of plant or leaf age and position and water scarce condition [8], on the other hand SCMR highly correlated with SLA and E [10-11]. Drought stress conditions increased chlorophyll density as well as transpiration efficiency. This result in the high leaf thickness in stress plants which, further causing higher chlorophyll content per unit area [12]. Drought stress majorly affects the food synthesizing machinery of leaves [13]. The interactions between moisture stress and morpho-physiological disorders with reduction of plant growth were previously studied [14-16]. Stomatal closure can be a common drought escaping response that allows plants to maintain water in their tissues [17]. In spite of severe drought conditions plants are able to sustain and perform gaseous exchange by intermittent opening of the stomata [18].

Water-use efficiency (WUE) measurement is a counter determinant of yield under stress condition [19-21]. Water use efficiency implies that rainfed plant production can be increased per unit water used resulting in more crop per drop. It implies maximal soil moisture capture for transpiration involves reduced non-stomatal transpiration and minimal water loss by soil evaporation [22]. In majority prevalent rainfed environmental conditions crop water scarcity develops during the reproductive growth stage consequently reducing yield[23-26].

Under drought stress conditions osmotic adjustment enables leaf turgor maintenance, therefore, irrespective of biomass production up to flowering, sustained WUE and *E* during the reproductive growth stage is crucial for reproductive success [27-29]. Water-limited condition resulted in a closure of stomata, which was accompanied by a marked decrease of net photosynthesis [30-32].

However, the photosynthetic efficiency of the Indian cluster bean cultivars together has not been evaluated yet and there is a strong need to characterize existing cultivars for various physiological traits and their association with yields for their better utilization in various environments. So, the present work emphasized to identify the the relatively better cluster bean variety against drought stress.

2. MATERIALS AND METHODS

The present study was conducted between May-July, 2014 in Rekulakunta, Regional Agricultural Research Station (RARS), Ananthapuramu (Lat 14˚ 41ˡ N and 77˚39ˡ E), Andhra Pradesh, India. The site is located in drought prone area with an annual rainfall of 640.2 mm per year and average temperature 34 ±5˚C.

Six cluster bean genotypes *i.e.* RGC-936, RGC-1025, HG-365, JJ-1, JG-2 and GLC-1031 were obtained from the Regional Agricultural Research Station (RARS), Rekulakunta, Ananthapuramu. Out of six genotypes RGC-936 and RGC-1025 were developed by Rajasthan, HG-365 is from Haryana whereas JJ-1 and JG-2 and GLC-1031 are developed by Gujarat.

2.1. Experimental Design

Six genotypes were planted in earthen pots (30×15×30) containing 15 kg of soil and sand mix in 1:3 ratio. Before sowing each pot was added with 200 g of FYM. Each pot accommodated with three plants. The entire experiment laid out in CRBD replicated three times.

2.2. Water Deficit Treatments

Plants were regularly irrigated at early hours approximately 8:30 AM and irrigation was continued up to 39 days. All the genotypes were subjected to different water regimes at 39 days after sowing (DAS). Soil moisture levels of 100%, 80%, 60%, 40% were maintained by weighing each pot on daily basis. The required level of the moisture range was reached on the 8th day of treatment (45 DAS) by decreasing the water supply progressively.

2.3. Leaf Exchange Gas Parameters

Leaf gas exchange parameters includes P_N [μmol m^{-2} s^{-1}], g_s [mol(H_2O) m^{-2} s^{-1}], *E* [mmol(H_2O) m^{-2} s^{-1}], C_i [μmol(CO_2) mol^{-1}] were measured on final day (8th day) of the treatment [33-34] by using *LICOR-6400XT* portable photosynthesis system (LI-COR Biosciences, INC Lincoln, Nebraska, USA) with a leaf chamber

(6×6 cm leaf area) of a temperature 34 ± 4 and RH 48˚C ± 10˚C . The measurement for gas exchange was recorded between 9:30 to 11:30 AM in medium portion of the 3rd leaf at 1600 PAR [μmol (photon) m^{-2} s^{-1}]with an ambient CO_2 concentration of 390 μmol mol^{-1}. Leaf chamber was not removed until the IRGA shows steady readings (Approximately 30sec). The gas exchange characteristics were calculated by default using the software of photosynthetic analyzer. The WUE*i* [μmol mol^{-1}] was calculated using values of P_N/g_s.

2.4. Relative Water Content

Leaf relative water content was calculated using standard methods [35]. Fully expanded leaves were cut from the main stem on the final day of the treatment at 11:30 AM. Fresh weight of the leaves was taken in 3 replicates and the leaves were subjected to rehydration in deionized water for 3 hours and turgid weight was measured. Later leaves were oven dried at 80˚C for 48 h. Based on the fresh weight (FW), dry weight (DW) and turgid weight (TW) of the leaves, the relative water content was measured using the formula RWC = (FW-DW/TW-DW) ×100.

2.5. SPAD and SLA

The SPAD Chlorophyll meter reading and SLA were recorded at 11:30 AM. SCMR was measured on the third fully expanded leaf by using *Minolta-502* SPAD meter (Tokyo, Japan). A maximum of 10 readings was taken for each leaf in triplicate (10×3 plants).

The second fully expanded leaf was detached and leaf area was measured using *LICOR 300C* leaf area meter (LICOR Inc, USA). The leaf samples were then oven dried at 80˚C for 72 h until to reach a constant weight. The SLA was recorded using the formula SLA= LA (cm^{-2})/ LDW (g).

2.6. Statistical Analysis

The data were subjected to ANOVA followed factorial experiment in 4×6 CRBD. The interaction between different water regimes and cluster bean genotypes were tabulated. The ANOVA was performed using AGRISTAT at 5% probably cut off. The Pearson correlations (r) between the leaf gas exchange variables, RWC, SPAD, and SLA were calculated using MINITAB16. The linear regression was analyzed in the present study using a probability value of 0.05% as a benchmark of significance.

3. Results

3.1. Gas Exchange Parameters

Gas exchange responses differed between all the varieties in all the levels of water stress. The average photosynthesis rate at fully hydrated conditions was 22.94 μmol m^{-2} s^{-1}. The mean percent reduction in net photosynthesis was shown 10.80%, 36.05%, 54.43% at 80, 60 and 40 percent of moisture availability respectively. The rate of photosynthesis was found to be significant in the varieties JJ-1 (25.45 μmol m^{-2} s^{-1}) and JG-2 (25.92 μmol m^{-2} s^{-1}) without water stress. At 80%, 60% and 40% water availability significant photosynthetic rate was observed in RGC-1025 (Figure 1A).RGC-1025 variety showed a maximum rate of g_s, C_i, E, RWC at 60%, and 40% water availability. These results indicated that photosynthesis was strongly associated with g_s, C_i, E, RWC parameters. In addition to RCG-1025.other two varieties RGC-936 and GLC-1031 shown significant photosynthetic rates (15.32 μmol m^{-2} s^{-1}) and (10.33 μmol m^{-2} s^{-1}) respectively at 60%, 40% water availability. Stomatal conductance values were measured in all the genotypes and the values were statically analyzed for their significance. The mean percent reduction in g_s was found to be more at 40% water availability (84.61%) (Figure 1B). At severe drought, the genotype RGC-1025 showed less percent of phosynthetic rate reduction at 60 (52.00%) and 80 percent (78.00%) water availability which is lower than total mean reduction percent *i.e* 84.61%. The data showed that JJ-1 (0.44 mol(H_2O) m^{-2} s^{-1}) and RGC-1025 (0.50 mol(H_2O) m^{-2} s^{-1}) were significant in controlled condition. At 80% moisture level HG-365 recorded more g_s (0.31 mol (H_2O) m^{-2} s^{-1}). At 60% water level varieties RGC-1025 (0.24 mol(H_2O) m^{-2} s^{-1}) and RGC-936 (0.23 mol (H_2O) m^{-2} s^{-1}) showed high g_s values. Internal CO_2 concentration reduced drastically in all the genotypes in all levels of water availability. Under fully hydrated conditions maximum C_i was ahown in RGC-936 (288.3 μmol (CO_2) mol^{-1}) (Figure 1C). With increased water stress (40% moisture level) only RGC-1025 recorded more C_i (223.96 μmol (CO_2) mol^{-1}) when compared to other varieties. Moreover, the percent reduction in C_i does not show much variability in RGC-1025 in all levels of moisture stress (14.22%, 18.27%, 22.32% at 80, 60, 40 percent water level respectively.). Transpiration followed similar trends as in net photosynthesis (P_N) and stomatal conductance (g_s). RGC-1025 shown maximum E in all levels of water stress (Figure 1D). Exceptionally, GLC-1031 and RGC-936 reported significant rate of transpiration at fully hydrated stage at 60%

water availabilityrespectively, other genotypes are not shown significant transpiration rates in any level of moisture. On the other side, a very interesting feature was observed *i.e,* WUE_i, SPAD and SLA did not show significant association with *E*. Maximum increase percent in intrinsic water use efficiency (P_N/g_s) was observed in 40% water availability (60.22%) over control plants (Figure 1E). Under normal water availability more WUE_i (71.80 μmol mol^{-1}) was observed in genotype JG-2. Inaddition to JG-2, higher WUE_i was observed in HG-365 (145.83 μmol mol^{-1}) and GLC-1031 (120.46 μmol mol^{-1}) at 60% water availability. At severe drought condition (40% water level) interestingly higher WUE_i values were observed in JJ-1 (167.84 μmol mol^{-1}) and JG-2 (184.58 μmol mol^{-1}). The significant WUE_i of JG-2 at 100% and 40% associated with P_N and WUE_i. On the other side, WUE_i (60% water level) of HG-365 also associated with RWC at 60% moisture availability. The instantaneous efficiency of water use efficiency (WUE_i) during the entire water deficit period exhibited an expressive increase with enhanced water stress and the mean WUE*i* was ranged from 3.02 mol (CO_2) mol $(H_2O)^{-1}$ to 4.33 mol (CO_2) mol $(H_2O)^{-1}$. The variety JG-2 recorded significant enhancement in WUE_i at 60% and 80% water level. This percent enhancement in *E* in JG-2 is 30.57% and 28.60% at 80 and 60 percent water availability. Inaddition to JG-2, reported maximum WUE_i in GLC-1031 and RGC-936 at 60% and 40% moisture level. WUE_i of JG-2 also showed association with RWC (80.33%) and SPAD (50.22 nmol cm^{-2}) at 60% water level. The genotype JG-2 recorded significant at 40% water level but not associated with RWC (Figure 1F). RGC-1025 was significant in controlled moisture level and 40% moisture level and it was associated with RWC. The enhanced WUE_i in JG-2 at 60% moisture level associated with RWC, SPAD and intrinsic water use efficiency at 40% water availability. Relative water content was more significant at all levels of moistures. The variety RGC-1025 showed significant RWC in all levels of water stress and it was associated with transpiration at all levels of moisture stress. The percent reduction in RWC does not vary much in RGC 1025, and at 40% water availability the mean percent reduction in RWC is 12.91% which is lesser than the mean percent reduction (13.97%) at severe drought stress. The RWC of the genotype RGC-936 found to be significant at 80% and 40% moisture stress levels whereas at 60% water availability and controlled conditions RGC-936 showed significant association with WUE_i, SPAD and and P_N, WUE_irespectively.

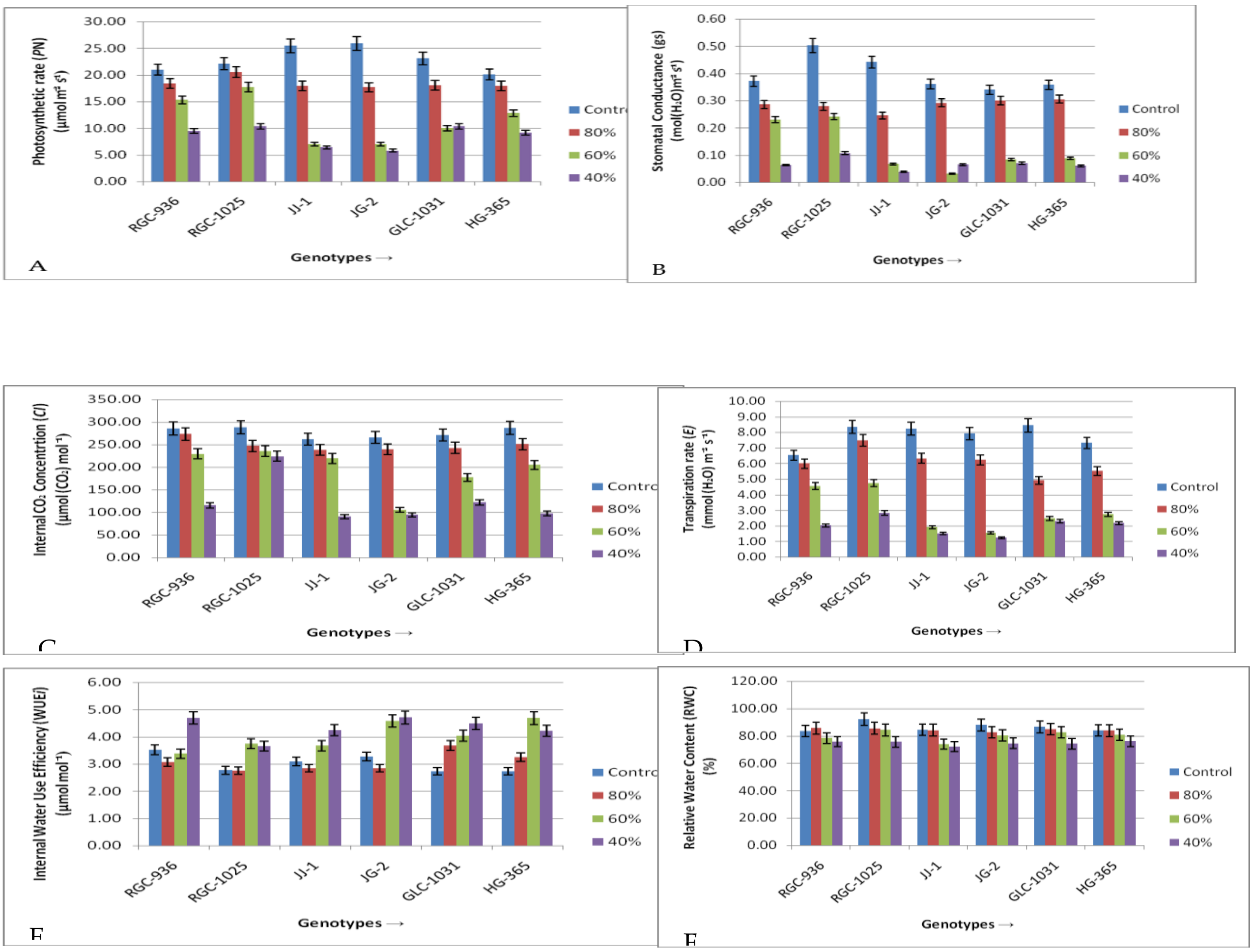


Figure 1 A-F. Fate of various physiological traits of genotyps under drought stress, A. Photosynthetic rate, B. stomatal conductance, C. Internal CO_2 concentration, D. Rate of transpiration, E. Internal water use efficiency, F. Relative water content. Cluster Bean genotypes: RGC-936, RGC-1025, HG-365, JJ-1, JG-2 and GLC-1031.

3.2. SPAD

SPAD meter readings were significant in two varieties RGC-1025 (51.32 nmol cm^{-2}), HG-365 (50.54 nmol cm^{-2}) at controlled conditions (Figure 2A). SPAD readings were associated with intercellular carbon dioxide and SLA values. In all levels of water stress, JJ-1 recorded significant values and it was found to be more in 30% water level (51.49 nmol cm^{-2}). At 80% moisture JJ-1 associated to SLA only. At 60% moisture stress JJ-1 and JG-2 were associated with intercellular carbon dioxide and JG-2 associated with WUE*i*. JJ-1 also associated with intrinsic water use efficiency at 40% water level.

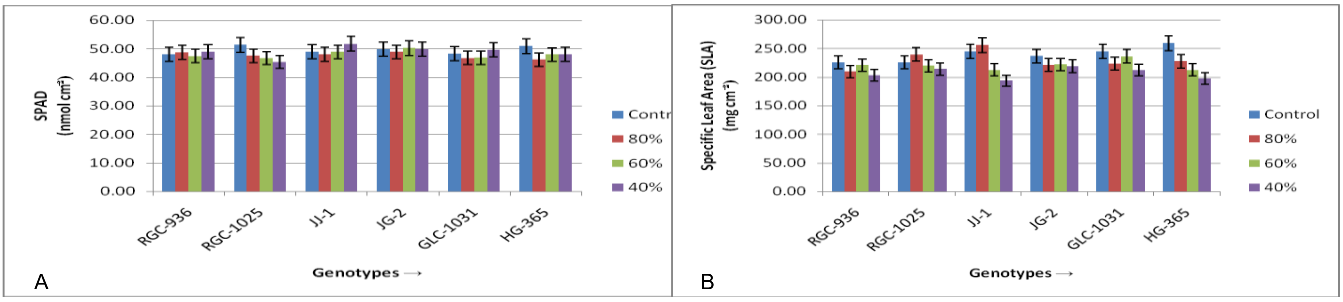


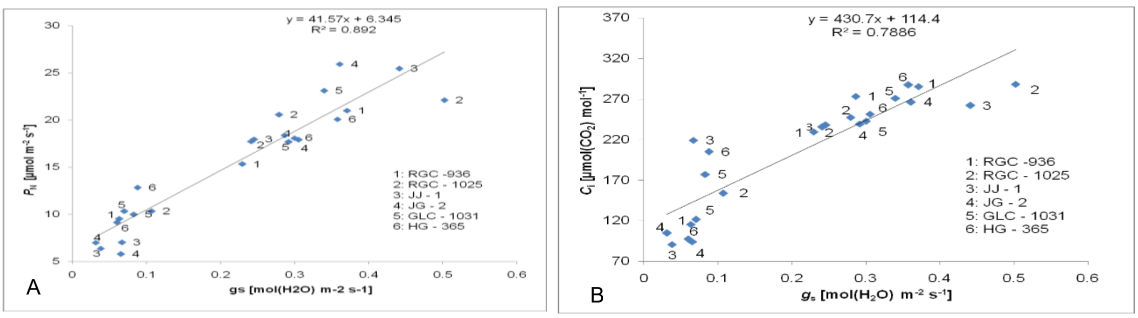
Figure 2 A. SPAD and B. Specific leaf area of genotypes under drought stress, Cluster Bean genotypes: RGC-936, RGC-1025, HG-365, JJ-1, JG-2 and GLC-1031.

3.3. SLA

The specific leaf weight values showed significant in all the controlled plants except JJ-1 and JG-2 (Figure 1H). In severe drought stress more SLA was expressed in JJ-1 (255.94 mg cm^{-2}) which is 16.94% more than the controlled plants. After JJ-1, RGC-1025 (239.64 mg cm^{-2}) and GLC-1031 (236.03 mg cm^{-2}) showed more SLA which is 5.82% increase in RGC-1025 over control and 31.62% decrease in GLC-1031 over control plants. At 80% water level JG-2 (221.24 mg cm^{-2}) and GLC (223.74 mg cm^{-2}) showed more SLA and at 60% water level JJ-1 (245.12 mg cm^{-2}) recorded significant SLA.

3.4. Correlation Analysis

Correlation analysis between the studied chacteristics under various water stress levels was calculated (Table 1). Net photosynthetic rate showed a strong positive association with g_s (0.923**) (Figure 3A) transpiration (0.962**) and SPAD (0.782**), P_N also showed strong association and with E at 40 % (0.933**) and RWC (0.768**) at 80% water level. P_N showed a negative association with C_i (-0.914**) under fully hydrated conditions. After C_i, photosynthetic rate showed strong association with WUE_i at 80% (-0.768**), 40% (0.780**) water level. With increased water stress level the negative correlation of P_N with C_i turn to positive and maximum positive association was observed in 60% water level (0.669**).



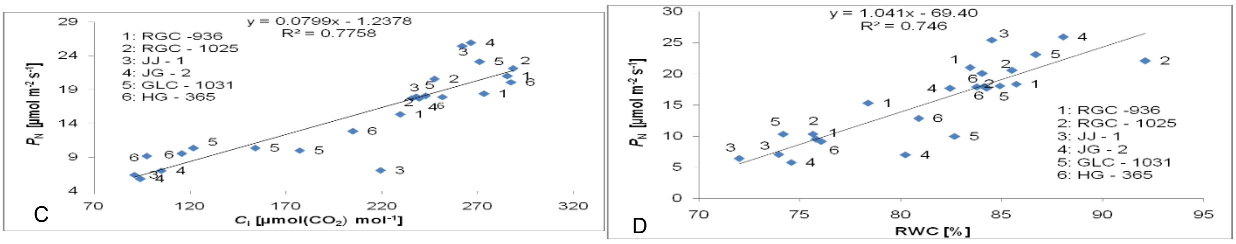


Figure 3. Regression analysis. A. between stomaal conductance and photosynthetic rate, B. between stomaal conductance and internal carbon dioxide concnetration, C. between internal carbon dioxide concnetration and photosynthetic rate under drought stress, D. between relative water content and photosynthetic rate under drought stress. Cluster Bean genotypes: RGC-936, RGC-1025, HG-365, JJ-1, JG-2 and GLC-1031.Stomatal conductance showed a strong association with C_i (0.735**) (Figure 3B) and E (0.991**) at 60% water availability. After $C_i$and E strong positive association between g_s and SLA was observed. In the present study, g_s expressed a strong negative association with WUE$_i$ at 100%, 60% and 40% water level. Stomatal conductance also exhibited a strong negative association with SPAD at 80%, 60%, 40% moisture level.

The C_i reported a strong association with P_N in all stress levels (80% = 0.568*; 60% = 0.669**; 40% = 0.603) (Figure 3C), E at 60% (0.718**), 40% (0.804**). Except E remaining all parameters showed negative association with C_i. Of them WUE$_i$ (-0.939**), WUE (-0.769**), SPAD (0.878**) recorded a strong negative association with C_i. Only SPAD showed a positive association with E at 100% (0.750**) and 80% (0.750**) water level, whereas WUE*i* (-0.684**), SPAD (0.845**) showed strong negative association with E. Only SPAD showed a strong positive association with WUE*i* at (0.680**) at 40% moisture, whereas WUE*i* exhibited a strong negative association with SPAD at 100% (-0.776**) and 80% (-0.780**) water availability. Specific leaf weight also expressed a strong negative association with WUE (-0.780**) at 100% water availability.

Relative water content recorded positive correlation with P_N (80% = 0.768**; 60% = 0.518*; 40% = 614**) (Figure 3D), SPAD (0.700**) at 40% moisture level and SLA (0.684**) at 60% water content. On the other side, RWC showed a significant negative association with SPAD 60% at water availability. SPAD recorded a positive relation with SLA at 100% water level and in remaining water levels SPAD does not show any significant the positive/negative association with SLA.

Table 1. Correlation analysis between various characteristics under various water stress levels

	Character Moisture level↓	Photosynthetic rate (µmol m^{-2} s^{-1})	Stomatal Conductance (mol(H_2O) m^{-2} s^{-1})	Internal CO_2 concentration (µmol (CO_2) mol^{-1})	Transpiration (mmol (H_2O) m^{-2} s^{-1})	Internal water use efficiency (µmol mol^{-1})	RWC (%)	SPAD (nmol cm^{-2})
Stomatal Conductance	100% 80% 60% 40%	0.084 0.152 0.923 0.518						
Internal CO_2 concentration	100% 80% 60% 40%	-0.914 0.568 0.669 0.603	0.179 0.517 0.735 0.903					
Transpiration	100% 80% 60% 40%	0.562 0.243 0.962 0.933	0.370 -0.331 0.991 0.684	-0.484 -0.484 0.718 0.804				
Internal water use efficiency	100% 80% 60% 40%	0.229 -0.304 -0.371 -0.386	-0.189 -0.250 -0.678 -0.584	-0.182 -0.182 -0.671 -0.769	-0.628 -0.628 -0.598 -0.684			
RWC	100% 80% 60% 40%	0.137 0.768 0.518 0.614	0.549 -0.000 0.276 0.495	0.209 0.209 -0.150 0.327	0.530 0.530 0.364 0.535	-0.317 -0.317 0.314 -0.063		

SPAD	100% 80% 60% 40%	0.245 0.216 -0.782 -0.699	0.344 -0.028 -0.743 -0.907	-0.133 -0.133 -0.599 -0.878	0.750 0.750 -0.792 -0.845	-0.776 -0.776 0.486 0.680	0.466 0.466 - 0.519 0.700	
Specific Leaf Area	100% 80% 60% 40%	-0.009 -0.198 -0.097 0.166	-0.210 -0.515 -0.154 0.691	-0.247 -0.589 -0.599 0.433	0.501 0.621 -0.116 0.150	-0.780 -0.438 0.220 0.128	- 0.261 - 0.277 0.684 0.355	0.582 0.352 0.211 - 0.441

DISCUSSION

Extreme drought conditions may affect the rate of photosynthesis. This reduction in photosynthesis may be due to the unavailability of Rubisco and its impaired activity [36-40]. The photosynthetic electron transport chain depends upon the availability of internal CO_2 concentration [41-42]. In present study variety, RGC-1025 showed maximum P_N may be due to its efficient maintenance of internal CO_2. On the other side dehydration due to severe water stress results in cell shrinkage and reduced cellular volume. This results increased cellular viscosity, a solute concentration which leads to the injury of cellular enzymes including the assimilatory mechanism [43-44]. But the variety RGC-1025 reported better RWC when compared to the other genotypes which tend to the record of high P_N values even at 40% water availability due to the efficient protecting mechanism of photosynthetic machinery. Effective transpiration, stomatal conductance mechanism of RGC-1025 in all levels of water stress tend to sustain better P_N rates. However, at high RWC most of the electrons participate in carboxylation process and very few are going to photorespiration. This mechanism found to be reversed under limited water avaialbility conditions[45].

A small reduction in RWC may reduce the P_N as low RWC tends to close the stomata highly effect the g_s coupled with C_i [46]. By changing more RWC, C_i, g_s, E at all levels of water stress RGC-1025 showing effective drought over resistance other genotypes.

Leaf cooling capacity has been given utmost priority for a better photosynthetic activity. Increased water stress level proportionally promotes the increased stomatal closure and reduced g_s. Stomatal closure tends to increase the leaf temperature and ultimately slow down and inefficient of Rubisco. The efficiency of RGC-1025 in making leaf environment cool at 60% and 40% water level implies that it has better protective mechanism over drought conditions. Inaddition to RGC-1025 the varieties RGC-936 and GLC-1031 shown higher P_N values may be because of their better g_s and E rates at 60% and 40% water level.

Stomata are the main channels to exchange gases like CO_2 and water. This condition implies that g_s is probably under stomatal regulation. Studies revealed that increased C_i in leaves results in partial closure of stomata [47-48]. In our study with increased water stress level, the internal C_i tend to decrease may be because of reduced stomatal conductance. But the variety RGC-1025 showed more g_s and C_i over even under severe drought conditions probably increased the P_N rate. On the other side P_N rate also depends on upon the rate of transpiration as transpiration is mainly driven by the water potential gradient between stomatal inner space and air-water potential [49-50]. With increased water stress all varieties of the present study showed reduced g_s by closing their stomata as they do not maintain water potential gradient of leaf and air. But the genotype RGC-1025 tried to maintain water potential gradient of leaf and air on par with atmospheric air results record of high P_N values even of 40% water availability.

In general intrinsic water use efficiency directly proportional to increased P_N. In present study varieties, JG-2 and JJ-1 recorded maximum WUE_i when compared to the other varieties. The same varieties also showed high WUE at 60% and 40% water availability. But the P_N rate was recorded low in these varieties at 60% and 40% water level. The displayed values of P_N, g_s, E in case of JJ-1 and JG-2 support their genetic inefficiency in performing carbon assimilation. In RGC-1025 even though it has recorded less WUE_i it tried to maintain g_s, E, along with RWC results in high P_N rate.

Assimilation rate generally positively associated with chlorophyll content per unit leaf area and hence increased SCMR and SLA will enhance P_N rate [51-53]. The chlorophyll density or SCMR values increased in moderate drought conditions because of the thickening of leaves in stress conditions but photosynthesis may not be increased. In the present study, JJ-1 recorded high SCMR of 60% and 40% level of water along with high SLA. But this genotype not able to express high P_N, g_s, and E. This is maybe because of its poor maintenance of water potential gradient. At the same time, RGC-1025 recorded high SLA, SCMR and hence recorded maximal P_N values in all levels of water stress.

CORRELATION ANALYSIS

Photosynthetic rate showed significant positive association with g_s in all the varieties of all stress levels. The leaf stomata plays role in gaseous change especially CO_2 indicating that g_s and P_N act together in performing normal assimilation [54-56]. The association between P_N and g_s is high in moderately stress condition showing that stomatal opening is more in moderate stress condition [57]. On the other hand, increased P_N positive association under moderate stress (60%= 0.923**) is due to the increased g_s which leads to the enhanced C_i association (0.669**) at 60% water availability. Increased stomatal opening results in higher E at (0.962**) at moderate stress level tends to reduce canopy temperature leads to the survivability of genotypes under increased water stress and heat due to dry soil conditions [58-60]. In our study, we also observed a strong association between RWC and P_N under all levels of stress. This adequate availability of leaf relative water trigger the Rubisco carboxylation and enhanced photosynthesis even depleted soil water level [61].

The strong negative association between WUE_i, WUE, with P_N, g_s, and E in all levels of water stress revealed that under severe water stress plants are unable to perform photosynthetic activity even though maximum g_s based on their poor genetic efficiency. The SCMR contents may directly relate to the enhanced P_N activity under increased water stress conditions. SCMR showed a strong positive association with P_N at 60% (0.782**) water availability. But at 40% water level it showed negative assimilation indicating that under severe water stress plants unable to perform photosynthesis due to reduced protein synthesis of photosynthetic machinery [62-63].

The SLA showed strong negative association with P_N, g_s, C_i at 100%, 80% and 60% water level. Whereas it showed a positive association with P_N, g_s, and C_i at 40% water availability. This positive association at severe water stress may be due to decreased SLA and pooling up of chloroplast in smaller leaf area [64-65].

CONCLUSIONS

Gas exchange parameters are found to be key factors in crop improvement pogramme of cluster bean under water stress. Understanding the contribution of various physiological traits in drought tolerance should pave the way to develop drought adopting clusterbean genotypes. Net photosynthetic rate (P_N) and stomatal conductance (g_s) showed strong association and with transpiration (E) at various water levels. But P_N reported a negative association with internal carbon dioxide (C_i) concentration under fully hydrated conditions. The C_i found to be associated strong with internal water use efficiency (WUE_i), and sotmatal conductance (g_s) at moderate and severe water stress levels where as gs associated negatively with SPAD at all water stress levels. Under fully hydrated conditions higher C_i was displayed in RGC-936. The variety RCG-1025 performed well and sustained a maximum rate of g_s, C_i, E, and RWC in moderate and severe water stress conditions when compared to other genotypes. The significant WUE_i of JG-2 associated with P_N and WUE*i*. The SPAD readings were associated with C_i and SLA. The SLA was varied significantly in all the controlled plants except JJ-1 and JG-2. In present study varieties JG-2 and JJ-1 recorded maximum WUE_i when compared to the other varieties. RGC-1025 recorded less WUE_i and tried to maintain g_s, E, along with RWC which results in high P_N rate, SLA, SCMR in all levels of water stress.

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