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Physiological and biochemical behavior in banana cultivars and hybrids under water deficit

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Water deficit is a major problem in banana grown under tropical and sub-tropical region climatic conditions. It affects plant growth and development and ultimately leads to a considerable bunch yield reduction or crop failure. Although the banana cultivars and hybrids are susceptible to water deficit, there is a marked genotypic variation in morphology of the plant and rooting pattern in banana in response to water deficit. The objective of this experiment was to investigate the possibility that the twelve banana cultivars and hybrids referred to as, Karpuravalli (ABB), Karpuravalli x PisangJajee, Saba (ABB), SannaChenkathali (AA), Poovan (AAB), Ney poovan (AB), Anaikomban (AA), Matti x Cultivar Rose, Matti (AA), Pisang Jajee x Matti, Matti x Anaikomban and Anaikomban x PisangJajee and coded as S_1, S_2, S_3, S_4, S_5, S_6, S_7, S_8, S_9, S_{10}, S_{11} and S_{12} might reveal cultivars specific diversity in the regulatory mechanisms underlying their different responses to water deficit with a view of establishing the most drought tolerant [Karpuravalli (ABB), Karpuravalli x PisangJajee, Saba (ABB)] banana cultivars and hybrids that can be grown under water deficit conditions in Tamil Nadu. The physiological and biochemical behavior in banana cultivars and hybrids subjected to water restriction were investigated during the 3rd, 5th, 7th and 9th month after planting (MAP). The parameters that were measured are the leaf relative water content, proline, total sugar, free amino acids and total soluble proteins. The experimental design carried out was at entirely split plot, with two water regimes considered as mail plot like stress (M1) and control (M2) and sub plot (S) taken as cultivars and hybrids. There was decrease in the leaf relative water content and soluble protein content in plants with decreased in water content. Results indicate that S_1 and S_2 has superior physiological traits under water deficit hence may be recommended for growing under water deficit conditions (50% available soil moisture) in tropical and subtropical regions.

Key words: Water deficit, banana, Relative water content (RWC), soluble protein, proline, free amino acid, total sugar.

INTRODUCTION

Water deficit is one of the most environmental stresses affecting agricultural productivity around the world and may result in considerable yield reductions (Boyer, 1985). Drought affects nearly all the plant growth processes; however, the stress response depends upon the intensity, rate, and duration of exposure and the stage of crop growth. Inhibition of leaf growth by water stress can be considered to be an adaptive response. Thus it limits leaf area production, eventually plants rate of transpiration (Lu and Neumann, 1998). Banana coming under tropical fruit and is the fourth largest fruit crop in the world. Although it has scores of definitions, it originates

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from a deficiency of precipitation over an extended period of time, usually a season or more. This deficiency results in water shortage for some activity, group, or environmental sector. Banana production and productivity is severely affected by various stresses like, drought, water and cold. Physiological responses to soil water deficit are the feature that is most likely to determine the response of the crop to irrigation. There are some evidence that roots are the primary sensors of water deficit in the soil, causing the observed physiological and biochemical perturbations in the stems and the decline in growth to be generally interconnected with changes in plant nutrition, carbon dioxide balance and water relations. The banana plants sensitivity to soil moisture stress, reflected in changes in reduced growth through reduced stomatal conductance and leaf size leads to reduction in photosynthetic pigments (Kallarackal et al., 1990) with increased leaf senescence (Turner, 1998). Bananas (*Musa* spp.) rarely attain their full genetic potential for yield due to limitations imposed by water ultimately limiting the plants photosynthesis.The water deficit is characterized by water losses that exceed the absorption rate and of this way it acts directly in the plant water relations (Costa et al., 2008), in which the plant damages depend on the intensity and the exposure period, besides promoting changes in the cell and the molecular pathways, as well as is reported accumulation of organic solutes as carbohydrates and proline (Lacerda et al., 2001), differential gene expression on nucleic acids as DNA and RNA (Casagrande et al., 2001), variation on the amount of photosynthetic pigments, mainly chlorophylls and carotenoids (Chandrasekar et al., 2000), in which the mechanism stomatal interferes on photosynthesis rates (Ribas-Carbo et al., 2005). The osmotic adjustment is considered an important mechanism developed by the plants to tolerate water deficiency (Costa, 1999), which promotes protection of the plant cell structures as membranes and chloroplasts (Martínez et al., 2004). It alsoavoids cell toxicity provoked by free radicals and maximizes water retention inside the cell (Hare et al., 1998), moreover it presents the possibility of using the total sugar as energy source under severe stress (Pimentel, 2004).The experiment aimed at evaluating the effects of the progressive water deficit, as well as to investigating the physiological and biochemical behavior in twelve banana cultivars and hybrids submitted to water restriction during the different growth stages.

MATERIALS AND METHODS

The experimental design was a split plot design with three replications. The main plots are, M_1 (control) with the soil pressure maintained from -0.69 to -6.00 bar, M_2 (water deficit) with the Soil pressure maintained from -0.69 to -14.00 bar. Soil pressure of -14.00 bar was reached at 30 days and measured by using soil moisture release curve and measured the soil moisture by using the pressure plate membrane apparatus (Table 1 and Figure 1). The sub plots considered as twelve banana cultivars and hybrids S_1: Karpuravalli (ABB), S_2: Karpuravalli x PisangJajee, S_3: Saba (ABB), S_4: SannaChenkathali (AA), S_5: Poovan (AAB), S_6: Ney poovan (AB), S_7: Anaikomban (AA),S_8: Matti x Cultivar Rose,S_9: Matti (AA),S_{10}: Pisang Jajee x Matti,S_{11}: Matti x Anaikomban and S_{12}: Anaikomban x PisangJajee were randomly distributed within the sub-plots in each of the drought stress treatments (main plots). The antioxidative enzymes were estimated during 3rd, 5th, 7th, 9th month after planting and at harvest stages of the crop.

Enzyme extractions and assays

Relative water content

Relative water content (RWC) was estimated according to the method of Weatherly (1950) and calculated in the leaves for each drought period. Samples (0.5 g) were saturated in 100 ml distilled water for 24 h at 4°C in the dark and their turgid weights were recorded. Then they were oven-dried at 65°C for 48 h and their dry weights were recorded. RWC was calculated as follows:

$$RWC\ (\%) = [(FW - DW) / (TW - DW)] \times 100,$$

Where, FW, DW, and TW are fresh weight, dry weight and turgid weight, respectively.

Soluble protein content

The content of soluble protein was estimated from the leaf samples following the method of Bradford (1974) and expressed as mg g^{-1} fresh weight.

Procedure: The leaf sample of 0.5 g was macerated with 10 ml of phosphate buffer (0.1 M, pH 7.0) using a pestle and mortar. The extract was centrifuged at 10000 rpm at 4°C for 20 min. 0.1 ml of supernatant was taken and 5 ml of dye mixture was added. The solution was mixed well and kept aside for 15 min. The colour intensity was recorded at 595 nm optical density.

Proline

Proline content of the leaf sample was estimated by the method of Bates et al. (1973) and expressed as µg g^{-1} of fresh weight.

Procedure: A fresh leaf sample of 0.5 g was macerated with 10 ml of aqueous sulphosalicylic acid (3%) using a pestle and mortar. The extract was centrifuged at 4000 rpm for 10 minutes. The supernatant solution of 2 ml was taken in a test tube and to this 2 ml of acid ninhydrin and 2 ml of glacial acetic acid was added.

The solution was kept in water bath for one hour at 100°C and it was cooled under tap water. After cooling, the solution was transferred into a separating funnel and 4 ml of toluene was added. The funnel was uniformly shacked for 30 s. Two different layers were formed. The colorless bottom layer was discarded and the upper pink color layer was collected. The optical density was recorded at 520 nm against blank as toluene.

Acid ninhydrin

2.5 g of ninhydrin was taken and mixed with 60 ml of glacial acetic acid and 40 ml of 6 M orthophosphoric acid. The solution was stirred well and slightly warmed in hot water bath until the content dissolved).

Free amino acids (FAA)

The free amino acids (FAA) of the leaf sample was estimated by the method proposed by Mishra and Dhillon (1981) and expressed as µg g^{-1}.

Procedure: A fresh leaf sample of 1 g was macerated with 10 ml of sodium phosphate buffer (0.2 M, pH 5.0) using a pestle and mortar. The extract was centrifuged at 10000 rpm at 4°C for 15 min. The supernatant solution of 0.1 ml was taken and 1 ml of ninhydrin solution was added and made upto 2 ml using distilled water and immediately kept in water bath for 20 min and allowed it for cooling at room temperature. After cooling, 5 ml of diluents solvent was added and allowed it to stand for 15 min. The intensity of purple color was noticed. The optical density was recorded at 570 nm against blank.

Diluents solution

Mix equal volume of water and n-propanol and this solution was used for enzyme analysis.

Ninhydrin solution

0.8 g of stannous chloride was dissolved in 500 ml of citrate buffer (0.2 M, pH 5.0). The solution was mixed in to 20 g of ninhydrin in 500 ml of methyl cellosolve (2 methoxy ethanol) and this solution was used for enzyme analysis.

Total sugars

The total sugar content of the leaf sample was estimated by the method of Nelson (1944) and Somogyi (1952) and expressed as mg g^{-1}.

RESULTS

Leaf relative water content

The leaf relative water content (LRWC) was reduced significantly (Table 2), as well as in the treatment under water deficiency had progressivefall of 79.2 to 68.7% in the LRWC during water deficit conditions respectively. Moreover, significant changes were showed within the stress treated cultivars and hybrids under waterdeficit, when compared with the control plot cultivars and hybrids. The highest leaf RWC was observed in Karpuravalli under the normal irrigated conditions with slight reduction was showed under water deficit. Matti, PisangJajee x Matti, Matti x Anaikomban and Anaikomban x PisangJajee cultivars and hybrids had highest reduction in leaf RWC under the water deficit, respectively. There was a high and positive correlation between RWC and proline water deficit conditions.

Soluble protein

The result on soluble protein content was affected by water deficit in all the cultivars and hybrids as well as the interaction of M at S and S at M were significant (Table 3). Water deficit reduced the soluble protein content banana cultivars and hybrids. Among the twelve cultivars and hybrids, Karpuravalli, Karpuravalli x PisangJajee, Saba, and Sannachenkathali had significant differences in soluble protein content under the main plot treatments. The highest soluble protein content was observed in Karpuravalli under the water deficit than the other cultivars and hybrids. The lowest soluble protein content was observed in Matti, PisangJajee x Matti, Matti x Anaikomban and Anaikomban x PisangJajee cultivars and hybrids under the water deficit, respectively. The positive correlation was showed between soluble protein contentand RWC under the water deficit conditions.

Proline

The proline level had significant changes, as well as in the plants under stress was showed total increase at 48% in free proline during the period of water deficit conditions (Table 4). Moreover, the cultivars of Karpuravallihad highest proline accumulation during water deficit conditions than the other cultivars and hybrids. The lowest poline accumulation was noticed in Matti, PisangJajee x Matti, Matti x Anaikomban and Anaikomban x PisangJajee cultivars and hybrids under the water deficit, respectively.

Free amino acids

The amino acids level were significant increase, in which the water deficit promotes total increase at 39% in amino acids level under the water deficit conditions (Table 5). Among the twelve cultivars and hybrids, Karpuravalli, Karpuravalli x PisangJajee, Saba, and Sannachenkathali had significant differences in free amino acid content under the main plot treatments. The highest free amino acid content was observed in Karpuravalli under the water deficit. The lowest free amino acid content was observed in Matti, PisangJajee x Matti, Matti x Anaikomban and Anaikomban x PisangJajeecultivars and hybrids during the water deficit, respectively.

Total sugar

The total soluble carbohydrate levels were progressively increased on the treatments control and stress, as well as the variance analysis reveals that occur significant difference among the treatments. In the twelve banana cultivars and hybrids submitted to stress was showed the increase at 30.36% (Table 6) during different growth stages due to water restriction, as well as great accumulation was showed in the period under water

Table 1. Calculated pressure from stress treatment and soil moisture content from regression equation.

Soil moisture content (%)	Pressure (bar)	ASM (%)
33.46	**-0.69**	**100.00**
31.32	-2.46	93.60
30.19	-3.39	90.23
29.18	-4.22	87.21
28.14	-5.08	84.10
27.09	-5.94	80.96
26.12	-6.74	78.06
25.29	-7.43	75.58
24.91	-7.74	74.45
24.32	-8.22	72.68
23.78	-8.67	71.07
23.40	-8.98	69.93
23.11	-9.22	69.07
22.86	-9.43	68.32
21.28	-10.73	63.60
20.83	-11.10	62.25
19.51	-12.19	58.31
19.30	-12.36	57.68
18.63	-12.91	55.68
18.11	-13.34	54.12
17.81	-13.59	53.23
17.52	-13.83	52.36
17.10	**-14.01**	**50.11**
16.72	-14.47	49.01
16.00	-15.08	47.82

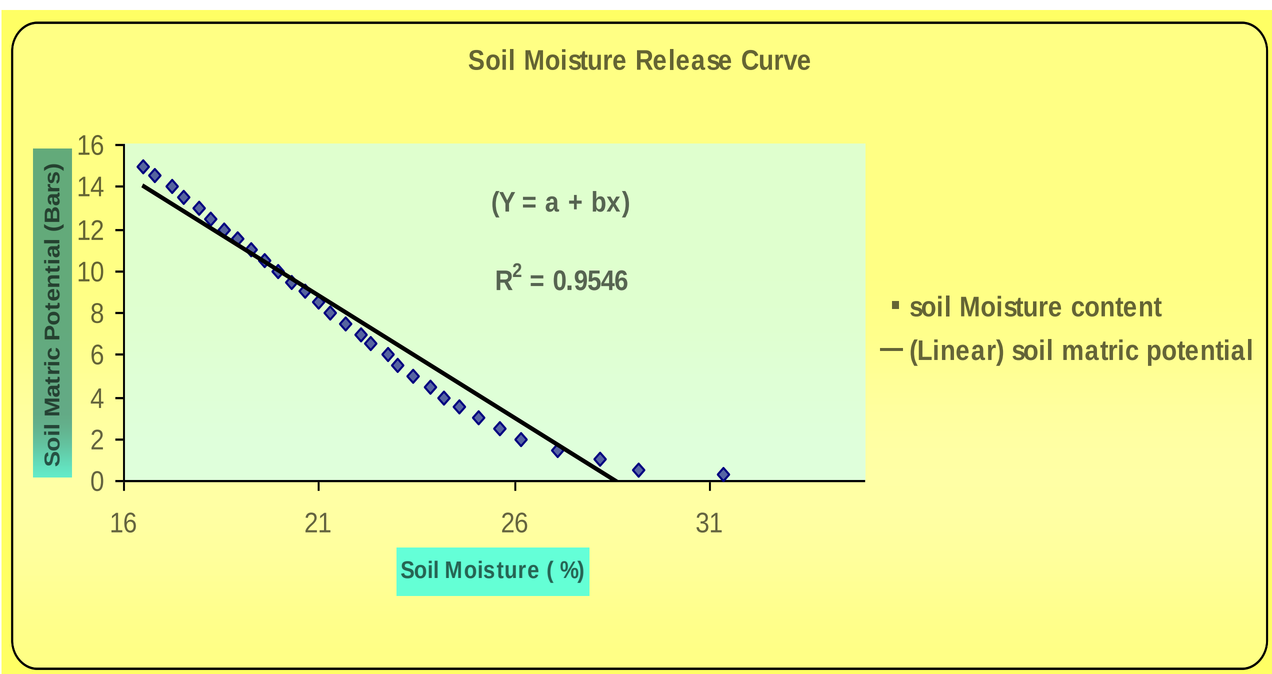

Figure 1. Pressure plate apparatus soil moisture release curve. Regression equation to find out pressure from soil moisture: [Y = a + bx]. Y = Pressure (bar); X = soil moisture content (%); ‘a’ = 28.26158; ‘b’ = - 0.8239.

Table 2. Effect of water stress on Relative Water Content (RWC: %) at different growth stages of banana cultivars and hybrids.

Treatments	3rd MAP	5th MAP	7th MAP	9th MAP	Harvest	Mean
Main plot						
M_1	75.4	77.1	78.2	73.1	72.9	75.35
M_2	65.9	67.6	68.7	63.6	63.4	65.80
Mean	70.64	72.34	73.44	68.34	68.14	70.58
SEd	0.42	0.43	0.48	0.41	0.41	
CD (P= 0.05)	1.83	1.87	2.06	1.77	1.76	
Sub plot						
S_1	81.0	82.7	83.8	78.7	78.5	80.92
S_2	80.3	82.0	83.1	78.0	77.8	80.22
S_3	77.4	79.1	80.2	75.1	74.9	77.32
S_4	76.9	78.6	79.7	74.6	74.4	76.82
S_5	74.1	75.8	76.9	71.8	71.6	74.02
S_6	73.4	75.1	76.2	71.1	70.9	73.32
S_7	69.7	71.4	72.5	67.4	67.2	69.62
S_8	68.0	69.7	70.8	65.7	65.5	67.92
S_9	63.5	65.2	66.3	61.2	61.0	63.47
S_{10}	61.4	63.1	64.2	59.1	58.9	61.37
S_{11}	61.1	62.8	63.9	58.8	58.6	61.07
S_{12}	60.9	62.6	63.7	58.6	58.4	60.87
Mean	70.64	72.34	73.44	68.34	68.14	70.58
SEd	0.90	0.92	0.93	0.87	0.87	
CD (P= 0.05)	1.82	1.86	1.88	1.77	1.76	
Interaction SEd						
M at S	1.29	1.32	1.35	1.25	1.25	
S at M	1.28	1.30	1.32	1.24	1.23	
CD (P= 0.05)						
M at S	2.93	3.00	3.12	2.84	2.83	
S at M	2.57	2.63	2.66	2.50	2.49	

deficiency.

DISCUSSION

Relative water content

Relative water content (RWC) is the appropriate measure of plant water status in terms of the physiological consequence of cellular water deficit. It was used instead of plant water potential as RWC referring to its relation with cell volume, which could accurately indicate the balance between absorbed water by plant and lost through transpiration. The banana plants are able to maintain their internal water status during drought by reducing radiation load and closing stomata (Thomas and Turner, 1998). The RWC was estimated in order to find out the plant water status of banana cultivars under water stress situations. Leaf RWC had a significant influence on photosynthesis, by reducing the net photosynthesis by more than 50% when RWC was less than 80%. As observed by Slatyer (1955), a reduction by 5% in RWC led to reduction in photosynthesis by 40 to 50%. Among the twelve cultivars, Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali had higher RWC content with 6% reduction over control. These findings were in agreement with the results of Thomas and Turner (1998), in which a positive correlation between RWC and gas exchange activities was observed and therefore, the reduction of RWC was found to cause a strong reduction in photosynthesis, transpiration and stomatal conductance. Gupta and Sing(1970) reported that drought resistant varieties showed consistently higher leaf water potential in their tissues than susceptible types under soil moisture deficit. In the present studies, cultivars like Matti, Matti x Anaikomban, Matti x cultivar rose and Pisangjajee x Matti, recorded lower RWC with higher reduction per cent in the range of

Table 3. Effect of water stress on foliage soluble protein (mg g^{-1})at different growth stages of banana cultivars and hybrids.

Treatments	3rd MAP	5th MAP	7th MAP	9th MAP	Harvest	Mean
Main plot						
M_1	10.7	10.8	11.2	9.6	5.5	9.58
M_2	8.7	8.8	9.9	8.2	4.6	8.03
Mean	9.67	9.81	10.54	8.93	5.06	8.80
SEd	0.049	0.049	0.053	0.048	0.024	
CD (P= 0.05)	0.211	0.214	0.229	0.208	0.106	
Sub plot						
S_1	11.8	11.9	12.7	11.8	6.1	10.85
S_2	10.9	9.4	10.6	10.9	5.0	9.36
S_3	10.3	9.1	9.9	9.4	4.9	8.71
S_4	8.8	9.4	9.9	7.3	5.7	8.23
S_5	9.7	10.5	11.5	6.9	5.0	8.69
S_6	8.4	10.9	11.3	7.7	5.8	8.82
S_7	9.4	9.8	9.8	7.8	4.6	8.25
S_8	9.4	8.9	10.4	8.9	4.8	8.47
S_9	10.2	8.2	10.0	9.5	4.8	8.55
S_{10}	10.1	9.5	9.3	9.6	4.6	8.61
S_{11}	9.0	10.5	10.5	9.0	4.8	8.75
S_{12}	8.2	9.6	10.7	8.5	4.7	8.35
Mean	9.67	9.81	10.54	8.93	5.06	8.80
SEd	0.129	0.132	0.134	0.123	0.067	
CD (P= 0.05)	0.261	0.267	0.270	0.249	0.136	
Interaction SEd						
M at S	0.183	0.186	0.188	0.174	0.094	
S at M	0.183	0.187	0.189	0.174	0.095	
CD (P= 0.05)						
M at S	0.405	0.405	0.409	0.382	0.205	
S at M	0.369	0.378	0.382	0.352	0.192	

Table 4. Effect of water stress on proline (µg g^{-1})at different growth stages of banana cultivars and hybrids.

Treatments	3rd MAP	5th MAP	7th MAP	9th MAP	Harvest	Mean
			Main plot			
M_1	73.6	75.3	79.4	71.3	71.1	74.15
M_2	91.9	93.4	98.3	89.6	89.4	92.52
Mean	82.76	84.33	88.86	80.46	80.26	83.33
SEd	2.00	2.01	2.01	1.98	1.98	
CD (P= 0.05)	8.60	8.64	8.67	8.55	8.54	
Sub plot						
S_1	95.8	96.7	98.4	93.5	93.3	95.55
S_2	95.1	96.1	97.8	92.8	92.6	94.90
S_3	92.2	93.9	94.9	89.9	89.7	92.14
S_4	91.7	93.4	94.4	89.4	89.2	91.64
S_5	84.9	86.6	87.6	82.6	82.4	84.83
S_6	84.2	85.9	86.9	81.9	81.7	84.13

Table 4. Contd.

S_7	80.5	82.2	83.2	78.2	78.0	80.43
S_8	78.8	80.5	83.4	76.5	76.3	79.11
S_9	74.2	75.9	82.1	71.9	71.7	75.18
S_{10}	72.1	73.8	84.6	69.8	69.6	74.00
S_{11}	71.8	73.5	85.3	69.5	69.3	73.90
S_{12}	71.6	73.3	87.5	69.3	69.1	74.18
Mean	82.76	84.33	88.86	80.46	80.26	83.33
SEd	0.88	0.90	0.91	0.85	0.85	
CD (P= 0.05)	1.78	1.82	1.84	1.73	1.72	
Interaction SEd						
M at S	2.33	2.35	2.36	2.30	2.30	
S at M	1.25	1.27	1.29	1.21	1.21	
CD (P= 0.05)						
M at S	8.62	8.67	8.69	8.57	8.55	
S at M	2.51	2.57	2.60	2.44	2.44	

Table 5. Effect of water stress on free amino acid (mg g^{-1}) at different growth stages of banana cultivars and hybrids.

Treatments	3rd MAP	5th MAP	7th MAP	9th MAP	Harvest	Mean
Main plot						
M_1	9.45	11.86	12.49	10.70	7.88	10.47
M_2	12.31	14.72	16.16	14.04	11.22	13.69
Mean	10.88	13.29	14.32	12.37	9.55	12.08
SEd	0.159	0.173	0.177	0.168	0.150	
CD (P= 0.05)	0.686	0.746	0.764	0.723	0.648	
Sub plot						
S_1	15.78	18.19	19.21	17.42	14.60	17.04
S_2	14.13	16.54	17.41	15.62	12.80	15.30
S_3	14.05	16.46	17.28	15.49	12.67	15.19
S_4	10.30	12.71	13.17	11.38	8.56	11.22
S_5	10.25	12.66	13.68	11.89	9.07	11.51
S_6	10.14	12.55	13.42	11.63	8.81	11.31
S_7	9.86	12.27	13.09	11.30	8.48	11.00
S_8	9.53	11.94	12.40	10.61	7.79	10.45
S_9	9.30	11.71	13.23	10.94	8.12	10.66
S_{10}	9.19	11.60	13.12	10.83	8.01	10.55
S_{11}	9.15	11.56	13.08	10.79	7.97	10.51
S_{12}	8.83	11.24	12.76	10.47	7.65	10.19
Mean	10.88	13.29	14.32	12.37	9.55	12.08
SEd	0.143	0.168	0.174	0.155	0.126	
CD (P= 0.05)	0.288	0.339	0.351	0.313	0.255	
Interaction SEd						
M at S	0.251	0.286	0.295	0.269	0.228	
S at M	0.202	0.237	0.246	0.219	0.179	
CD (P= 0.05)						
M at S	0.737	0.817	0.839	0.783	0.688	
S at M	0.408	0.478	0.497	0.442	0.361	

Table 6. Effect of water stress on leaf total sugar (mg g^{-1}) at different growth stages of banana cultivars and hybrids.

Treatments	3rd MAP	5th MAP	7th MAP	9th MAP	Harvest	Mean
			Main plot			
M_1	6.03	7.07	7.49	7.39	5.92	6.78
M_2	6.82	6.42	8.78	8.68	7.13	7.56
Mean	6.42	6.74	8.13	8.03	6.52	7.17
SEd	0.086	0.055	0.119	0.095	0.046	
CD (P= 0.05)	0.370	0.240	0.512	0.410	0.199	
			Sub plot			
S_1	11.57	11.82	13.89	13.79	12.32	12.68
S_2	10.45	10.65	12.77	12.67	11.20	11.55
S_3	9.38	9.63	11.70	11.60	10.13	10.49
S_4	7.31	7.66	9.53	9.43	7.96	8.38
S_5	5.43	5.78	7.10	7.00	5.28	6.12
S_6	5.11	5.46	6.68	6.58	5.01	5.77
S_7	4.82	5.17	6.29	6.19	4.67	5.43
S_8	4.76	5.11	6.28	6.18	4.66	5.40
S_9	4.73	5.08	6.00	5.90	4.43	5.23
S_{10}	4.71	5.06	5.98	5.88	4.41	5.21
S_{11}	4.54	4.89	5.81	5.71	4.24	5.04
S_{12}	4.25	4.60	5.52	5.42	3.95	4.75
Mean	6.42	6.74	8.13	8.03	6.52	7.17
SEd	0.113	0.126	0.126	0.122	0.072	
CD (P= 0.05)	0.229	0.255	0.256	0.247	0.146	
Interaction SEd						
M at S	0.176	0.180	0.204	0.196	0.205	
S at M	0.160	0.179	0.173	0.179	0.172	
CD (P= 0.05)						
M at S	0.451	0.403	0.570	0.502	0.571	
S at M	0.324	0.361	0.349	0.361	0.348	

22 to 24% reduction when compared to control. Similarly in banana plants, Thomas and Turner (1998), found that, a major decrease of soil moisture hardly reduced the leaf RWC. The early reduction of stomatal conductance and the minor diminution of leaf RWC could indicate that the banana plants showed a drought avoidance mechanism to maintain a favorable plant water status involving stomatal closure in response to water stress (Turner and Lahav, 1983).

Soluble protein content

The soluble protein content of the leaf, being a measure of RuBP carboxylase activity was considered as an index for photosynthetic efficiency. There were reports that RuBP-case enzyme forms nearly 80% of the soluble proteins in leaves of many plants (Joseph et al., 1981). Diethelm and Shibles (1989) opined that the RUBISCO content per unit leaf area was positively correlated with that of soluble protein content of the leaf. Soluble protein content was estimated in order to find out the photosynthetic capacity of banana cultivars under water deficit situations. In the present study, water deficit caused a significant reduction in soluble protein content of leaves of all the banana cultivars. Among the cultivars studied, Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali maintained higher soluble protein content with 9% reduction over control, followed by cultivars of Poovan, Ney Poovan, Anaikomban and Anaikomban x Pisangjajee with 15% reduction. However, the cultivars of Matti, Matti x Anaikomban, Matti x cultivar rose and Pisangjajee x Matti recorded the lowest soluble protein content with higher reduction of 19% than control. The mechanism of reduction in soluble protein content due to water deficit by reduction in RUBISCO enzyme activity leads to lower CO_2 assimilation. These findings were in accordance with the results of Wahad et al. (2000) who observed a significant reduction in soluble protein content of banana plants grown under drought stress conditions. Besides these results, Martignone et al. (1987) observed that in soybean soluble protein content was the first nitrogenous compound affected under stress conditions, which at severity got denatured and lost the activity. It was further explained that soluble protein, world's most abundant protein containing the enzyme RUBISCO, is involved in CO_2 assimilation; therefore, the reduction in soluble protein might have a direct adverse effect on photosynthesis. Hsiao (1974) reported that the decrease in the protein level in water stressed plants could be attributed to decrease in protein synthesis, the decreased availability of amino acids and denaturation of the enzymes involved in amino acid and protein synthesis, perhaps at the ribosomal level.

Proline

Proline accumulation is an universal response of plants to various stresses. Proline acts as an osmolyte and helps the plants to maintain tissue water potential under all kinds of stresses. Proline, as an osmoprotectant, is largely confined to the cytoplasm and is mostly absent from the vacuole (Mc Neil et al., 1999). It plays a key role in the cytoplasm as a scavenger of free radicals as well as a mediator in osmotic adjustment and also increases the solubility of sparingly soluble proteins (Caplan et al., 1990; Saradhi et al., 1995). Shen et al. (1990) advocated that water stress enhanced the accumulation of proline in many plant species and it might function as a source of solute for intercellular osmotic adjustment under water stress. Stewart (1978) suggested that proline might severe as a storage compounds for reduced carbon and nitrogen during stress. Proline might regulate the osmotic balance of the cell thus relieving the negative effect of stress (Reddy et al., 2004). In the present study also, cultivars like Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali had higher amount of proline accumulation particularly at 7th MAP followed by Poovan, Ney Poovan, Anaikomban and Anaikomban x Pisangjajee than cultivars of Matti, Matti x Anaikomban, Matti x cultivar rose and Pisangjajee x Matti. These findings are further supported by the results of Mohd et al. (2004) in banana, which explained that the enhancement in free proline content could occur either due to *'de novo'* synthesis of proline or breakdown of proline-rich protein or shift in metabolism. According to Karamanos et al. (1983), there are three main reasons for the accumulation of proline in stressed leaves. The first and main component for this accumulation is the stimulation of proline synthesis from glutamic acid. The second is an inhibition of proline oxidation to other soluble compounds, and the third an inhibition of protein synthesis. The metabolic conversion of glutamic acid to proline and thereafter to other products via oxidation occurs readily in turgid leaves of barley and is stimulated by higher levels of proline.

Free amino acids

Accumulation of free amino acids is also significant under water stress and may be due to induced hydrolysis of proteins as reported in crops like *Arachis hypogeae* and *Vicia faba* (Purushotham et al., 1998; ElTayab, 2006). Jones et al. (1981) demonstrated the contribution of solutes in the osmotic adjustment during moderate stress levels than severe stress level. As observed in the present studies, cultivars of Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali registered 22% increase in FAA content than the control. These findings are in accordance with Stewart and Larher (1980) found an accumulation of free amino acids in the presence of water deficit leading to a dynamic adjustment of nitrogen metabolism. The increase in free amino acids could contribute to the tolerance of the plant to water deficit through an increase in osmotic potential or as a reserve of nitrogen principally for the synthesis of specific enzymes (Navari-Izzo et al., 1990). Kraus et al. (1995) reported that increased accumulation of FAA during stress conditions could be an indicator of the adaptive nature of the coconut palms to cope up with the adverse conditions during summer months. Once water deficit is established, the level of free amino acids in plants increased under moderate stress and severe stress conditions (Jones et al., 1981). The increase in aspatate, glutamate, proline, alanine and valine compound due to increase in free amino acids content in stressed leaves could help maintain energy fluxes of the chloroplast (Ashraf, 2004).

Leaf total sugar

Response of plants to abiotic stresses is reflected by a change in their total soluble sugars accumulation (Bruggemann et al., 1994; Conocono et al., 1998; Kumar, 2000). In the present study also, the cultivars like Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali recorded about 25% increase in leaf total sugar content under stress condition over the control at 7th MAP. Besides these results, Bruggemann et al. (1994) found significant increase in the total soluble sugar concentration in flag leaf under abiotic stress condition indicating that under abiotic stress, partitioning or translocation of soluble carbohydrates was adversely affected leading to poor utilization of assimilates in the sink. Other cultivars of Poovan, Ney Poovan, Anaikomban and Anaikomban x Pisangjajee also exhibited a change in total sugar content under water deficit condition with 17 to 20% increase over control. Similarly, a slight increase in total sugar content of 8 to

9% was also showed by Matti, Matti x Anaikomban, Matti x cultivar rose and Pisangjajee x Matti under water deficit condition over control. Waters et al. (1991) observed a noticeable decline in transport of sugars from shoot to root and accumulation of total sugar content in maize roots under oxidative stress.

Conclusion

Plants respond to drought stress through alteration in physiological and biochemical processes. Our results showed that the activities of proline, free amino acid and total sugar content were increased under the water deficit condition with reduction in RWC and soluble protein content. The banana cultivars and hybrids of Karpuravalli, Karpuravalli x Pisangjajee, Saba and Sannachenkathali with highest proline, free amino acid and total sugar content under water deficit. The findings of this research also showed that the proline, free amino acid and total sugar content content can be used as a drought tolerance index to selection tolerant genotypes under water deficit conditions in banana cultivars and hybrids.

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REFERENCES

Ashraf M (2004). Some important physiological selection criteria for salt tolerance in plants. Flora 199:361–376.

Bates LB, Waldren RP, Teare ID (1973). Rapid determination of free proline for water stress studies. Plant Soil 39:205- 207.

Boyer JS (1985). Water transport. Ann. Rev. Plant Physiol. 36:473-516.

Bruggemann W, Klauck S, Maas-Kantel K (1994). Long term chilling of young tomato plants under low light in lycopersicon esculentum Mill and L. Peruvianum Mill. Planta 194:160-168.

Caplan A, Claes B, Dekeyser R, Van Montagu M (1990). Salinity and drought stress in rice. In: The Impact of Biotechnology in Agriculture. Ed. R.S. Sangwan and B.S. SangwanNorrea.Kluwer Academic, Dordrecht. pp. 391-402.

Casagrande EC, Farias JRB, Neumaier N, Oya T, Pedroso J, Martins PK, Breton MC, Nepamuceno AL (2001) Differential gene expression in soybean during water deficit. Rev. Bras. Fisiol. Veg. 13:168-184

Chandrasekar V, Sairam RK, Srivastava GC (2000). Physiological and biochemical responses of hexaploid and tetraploid wheat to drought stress. J. Agron. Crop Sci. 185:219-227.

Conocono EA, Egdane JA, Setter TL (1998).Estimation of canopy photosynthesis in rice by means of daily increases in leaf carbohydrate concentrations. Crop Sci. 38: 987-995.

Costa RCL, Lobato AKS, Oliveira Neto CF, SantosFilho BG, Cruz FJR, Laughinghouse IV HD (2008). Biochemical and physiological behaviorof Vignaunguiculata (L.) Walp.under waterstress during the vegetative phase. Asian J. Plant Sci. 7:44-49

Costa RCL (1999) Nitrogen assimilation and osmotic adjustment in nodulated plants of bean [Vigna unguiculata (L.) Walp] under water stress, Ph.D Thesis. Universidade Federal do Ceará,

Diethelm R, Shibles R (1989). Relationship of enhanced sink demand with photosynthesis and amount and activity of Ribulose-1,5biphosphate carboxylase in soybean leaves. J. Plant. Physiol. 134:70-74.

Gupta SK, Singh TA (1970). Effect of some solute concentration of germination of some oilseeds. Mysore J. Agric. Sci. 4:221-223.

Hare PD, Cress WA, Staden JV (1998). Proline Synthesis and Degradation: a Model System for Elucidating Stress-related signal Transduction. J. Exp. Bot. 50:413-414.

HsiaoTC (1974). Plant responses to water deficit.Ann. Rev. Plant Physiol. 24:519:570.

Jones HG, Luton MT, Higgs KH, Hamer PJ (1981). Experimental control of water status in an apple orchard. J. Hort. Sci. 58:301-316.

Joseph MC, Randall DD, Nelson CJ (1981). Photosynthesis in polyploid tall fescue. II. Photosynthesis and RUBP case of polyploid tall fescue. Plant Physiol. 68:894-898.

Kallarackal J, Milburn JA, Baker DA (1990). Water relations of the banana. III effects of controlled water stress on water potential, transpiration, photosynthesis and leaf growth. Austr. J. Plant Physiol. (17):79-90.

Karamanos A, Drossopoulos JB, Niavis CA (1983). Free proline accumulation during development of two wheat cultivars with water stress. J. Agric. Sci. 100:429-439.

Kraus TE, Mckersie BD, Fletcher RA (1995). Paclobutrazole induced tolerance of wheat leaves to paraquat may involve antioxidant enzyme activity. J. Plant Physiol. 145:570– 576.

Kumar P (2000). Physiobiochemical changes caused by leaf rust in flag leaves of wheat. J. Bot. Soc. 79:169-173.

Lacerda CF, Cambraia J, Cano MAO, Ruiz HA (2001). Plant growth and solute accumulation anddistribution in two sorghum genotypes underNaCl stress. Brazilian J. Plant Physiol. 13:270-284.

Lu Z, Neumann PM (1998).Water-stressed maize, barley, and rice seedlings show species diversity in mechanisms of leaf growth inhibition. J. Exp. Bot. 49:1945–1952.

Martignone RA, Guiamot JJ, Nakayama F (1987). Nitrogen partitioning and leaf senescence in soybean as related to nitrogen supply. Field Crops Res. 17:17-20.

Martínez JP, Lutts S, Schanck A, Bajji M, Kinet JM (2004). Is osmotic adjustment required for waterstress resistance in the Mediterranean shrubAtriplexhalimus L. J. Plant Physiol. 161:1041-1051

Mc Neil SD, Nuccio ML, Hanson AD (1999). Betaines and related osmoprotectants - Targets for metabolic engineering of stress resistance. Plant Physiol. 120:945-949.

Mishra KA, Dhillon BS (1981). Ribonucleic acid, proteins, phenols and amino acids in the leaves in relation to fruit bud differentiation in mango. Indian J. Agric. Sci. 51:447-449

Mohd RI, Mohd KY, Mohamad HA (2004). Growth and physiological process of banana (Musa spp.) plants to soil water deficits. P. 343.

Navari-Izzo, F. Quartacci MF, Izzo R (1990). Water stress induced changes in protein and free amino acids in field grown maize and sunflower. Plant Physiol. Biochem. 28:531-537.

Nelson N (1944). A Photometric Adaptation of the Somogyi Method for the Determination of Glucose. J. Biol. Chem. 153:375-380.

Pimentel C (2004). Water relations in two hybridsof corn under two cycles of water stress. Pesq. Agropec.Bras. 34:2021-2027.

Purushotham MG, Patil TG, Prasad SN (1998). Development of in vitro PEG stress tolerant cell lines in two groundnut genotypes. Indian J. Plant Physiol. 3:49-51.

Reddy AR, Raghavendra AS, MadhavaRao KV (2004). Photooxidative stress; physiology and molecular biology of stress tolerance in plants. Springer, the Netherlands pp. 157-185.

Ribas-Carbo M, Taylor NL, Giles L, Busquets S,Finnegan PM, Day DA, Lambers H, Medrano H,Berry JA, Flexas J (2005). Effects of water stresson respiration in soybean leaves. Plant Physiol.139:466-473

Saradhi PP, AlianArora S, Prasad KVSK (1995). Proline accumulates in plants exposed to UV radiation and protects them against UV induced perioxidation. Biochem.Biophys. Res. Commun. 209:1-5.

Shen XY, Dai JY, Hu AC, Gu WL, . He RY, Sheng B (1990). Studies on physiological effect of Brassinosteroid as drought resistance

in maize. J.Shenyang Agri. Univ. 21(3):191-195.
Slatyer RO (1955). Modifications to the relative turgidity techniques with notes on its significance as an index of the internal water status of leaves. Austr. J. Agric. Res. 61:365-377.
Somogyi (1952). Notes on Sugar Estimation. J. Biol. Chem. 175:19-23.
Stewart CR, Larher F (1980). Accumulation of amino acids and related compounds in relation to environmental stress. The biochemistry of plants: a comprehensive treatise, New York: Academic Press. .5:609-635.
Stewart CR (1978). Rate of carbohydrates in proline accumulation in wilted barley leaves. Plant Physiol. 61:775-778.
Thomas DS, Turner DW (1998). Leaf gas exchange of droughted and irrigated banana cv. Williams (Musa spp.) growing in hot, arid conditions. J. Hort. Sci. Biotechnol. 73:419-429.
Turner DW (1998). The impact of environmental factors on the development and productivity of bananas and plantains.In Proceedings of the 13th ACORBAT meeting, Guayaquil, Ecuador, pp. 635-663 (Ed. L. H. Arizaga). Ecuador, CONABAN.
Turner DW, Lahav E (1983). The growth of banana plants in relation to temperature. Austr. J. Plant Physiol. 10:43-53.
Wahad Z, ZulkifliHj, Suzita S (2000). Proceedings of the first national banana seminar at AwanaGenting and country resort. UPM: Serdang (mys). P. 343.
Waters I, Morrell S, Greenway H, Colmer TD (1991). Effect of anoxia on wheat seedling II. Influence of oxygen supply prior to anoxia, alcoholic fermentation and sugar level. J. Exp. Bot. 42:1437-1447,
Weatherly PE (1950). Studies in the water relations of cotton. 1. The field measurement of water deficits in leaves. New Phytol. 49:81-97.