

Full Length Research Paper

Screening of low temperature tolerance on cassava genotypes according to stomatal conductances

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8 cassava genotypes were evaluated under growth chambers (15/10, 20/15, 25/20 and 32/22°C day/night temperature respectively) and field conditions (Ibadan: 27B ± 5°C and Jos plateau: 18 ± 5°C) from 1993 to 1996. In a growth chamber study, none of the tested genotypes survived after 3 week at 15/10°C, low temperature regimes by 20/15 and 25/20°C had significantly ($P < 0.05$) reduced leaf stomatal conductances compared to high temperature, 32/22°C. The abaxial stomatal conductance ranged from 0.13 to 0.63 mol m⁻¹ s⁻¹. The total dry biomass per plant was higher at ambient than at lower temperature (20/15°C). In a field study, stomatal conductances were significantly higher at Ibadan compared to Jos plateau. The leaf stomatal conductance ranged from 5.6 to 21.0 mol m⁻¹ s⁻¹. Genotypic variations were observed by stomatal conductances depending on temperature regimes and locations. At 20/15°C and Jos plateau, TMS 91934, Danwaru, TMS 30572 and TME1 had the highest leaf stomatal conductances. This result showed that high stomatal conductances contributed to the tolerance of TMS 91934, TMS 30572 and TME1 to low temperatures, these genotypes had the highest total dry biomass compared to other evaluated genotypes. The correlation of dry biomass with stomatal conductances ($r = 0.77$, $n = 8$) suggest that high stomatal conductances are important physiological characteristics of cassava genotypes tolerant to low temperatures.

Key words: Low temperature, stomatal, tolerance, cassava genotypes.

INTRODUCTION

Cassava is a staple food for over 800 million people around the world (CIAT, 1993; Nweke, 1996). Cassava is a chilling intolerant plant (Cock, 1985; Akparobi et al., 2002). Exposing chilling intolerant plant to low temperature results in the inhibition of cellular activities (Levitt, 1980). The regulation of photosynthesis activity may be necessary adaptation enabling stress resistant plants to avoid photodamage during exposure to low temperatures (Levitt, 1980). Also, there is increasing understanding that the effects of stress on photosynthetic rates can be partially attributed to stomatal function (Furbank and Badger, 1983; El-Sharkawy and Cock, 1990).

Stomata control the diffusive transfer of water vapour and carbon dioxide between the leaf and ambient air (Farquhar and Sharkey, 1982). The aerial environment affects stomatal aperture (Levitt, 1980). High temperatures up to 30°C tend to open stomata of cassava (Bueno, 1986; El-sharkway and Cock, 1990), while temperatures below 20°C close stomata of cassava plants (Ekanayake et al., 1997; Akparobi et al, 2002). Stomata response of plants to temperature is strongly influenced

by many factors of which internal plant water status and water vapour pressure deficit between leaf and surrounding air are particularly important (Berry and Bjorkman, 1980).

Considerable evidence has recently accumulated which indicates that measurements of photosynthetic rates are highly dependent upon the temperature conditions (El-Sharkway and Cock, 1990). Several workers have shown that plants that are subjected to low temperatures exhibit a reduction in net photosynthetic rate (Kramer, 1989; Berry and Bjorkman, 1980; Bhagasari, 1994). It has been observed that reduction in temperature causes a general decline in the rate of the dark reactions of photosynthesis (Running and Read, 1980).

The study of the diffusive resistances can yield information about the resistances to CO₂ exchange (Grantz and Meinzer, 1989; Bhagasari, 1994). However, in cassava scanty information is available on using stomatal conductances for screening cassava low temperature tolerance. Thus, the aim of this study is to screen cassava genotypes for low temperature tolerance using stomatal con-

ductances.

MATERIALS AND METHODS

Growth chamber study

Plant materials

8 cassava genotypes (5 improved IITA genotypes: TMS 30555, TMS 91934, TMS 4(2)1425, TMS 30001 and TMS 30572, one local landrace from southern part of Nigeria, TME1, 2 local landraces adapted to mid-altitudes of Jos plateau, Danduala and Danwaru) were grown in growth chambers. They were selected for this work on the basis of their origin, yielding ability and broad adaptation to several agroecological zones.

Growth facilities

Cassava plants were raised in growth chambers (Model Li-E15, convirons, controlled environments Ltd; Winning, Manitoba, Canada) at IITA, Ibadan. In each chamber, a 12 h daylength and day/night temperature regimes of 25/20, 20/15 and 15/10°C, were maintained. Overhead light was supplied in each growth chamber by a combination of 12 fluorescent and 16 incandescent lamps. The average photosynthetic photon flux (400 - 700 nm) was 10 - 15 mmol m⁻² s⁻¹. The relative humidity in each growth chamber ranged between 65 and 80%. The plants were grown in plastic pots (0.45 m diameter, 0.50 m deep) containing topsoil (sandy loam, pH 5.9 - 6.7). The soil used as potting medium is classified as oxic paleustalf, Alagba soil series (Greenland, 1981).

Plant culture

Cassava stem cuttings of 0.20 m length with 6 nodes were obtained from 12 months old mother plants at the middle part of stem and were planted in the plastic pots. Cuttings were planted inclined at 40° to the horizontal and watered 3 times a week. These cuttings were raised under natural conditions for 4 weeks until the cuttings were sprouted. The average maximum and minimum temperature and relative humidity during this period were 28 - 32, 21 - 24°C and 65 - 85%, respectively. After 1 month, sprouted plants were transferred to convirons set at 25/20, 20/15 and 15/10°C day/night temperatures respectively, while other plants remained outside the conviron area under ambient conditions (32/22°C). Throughout this experiment, plants in each pot were regularly supplied with water to field capacity (400 ml per pot) and were hand weeded. Whenever necessary 0.2% Perfekthion (Dimethoate 400 g/L E. C.) was applied to control mealy bug. This experiment was a factorial arranged in a completely randomized design in 3 replicates.

The field study

These experiments were conducted at Jos plateau (mid-altitude) and Ibadan (forest-savanna transition zone). 2 experiments were conducted during 1994/95 and 1995/96 crop seasons at international institute of tropical agriculture (IITA), Ibadan and national root crop research institute field stations (Vom and Heipang) in Jos plateau. These 2 locations represent contrasting agroecological zones: Ibadan (altitude: 210 m above sea level (masl), relative humidity: 65 - 90%, latitude: 4°46'N, longitude: 2°34'E, temperature: 28 ± 6°C, rainfall: 1545 mm) and Jos plateau (Vom: altitude: 1280 masl, latitude: 9°55'N, longitude: 9°E, relative humidity: 55 - 58%, rainfall: 1099 mm, temperature: 18 ± 5°C, Heipang: altitude: 1290 masl, latitude: 9°38'N, longitude: 8°9'E, relative humidity: 60 - 85%, temperature: 18 ± 6°C, rainfall: 1153 mm).

Similar cassava genotypes for growth chamber were used in the field study. The experiments were set up in each location in a completely randomized block design with 3 replications. Each plot had 6 rows, 10 m long. Spacing was 1 m between rows and 0.8 m within a row. Each plot contained 72 plants. Fields were kept free of weeds by regular hand-weeding. The soil at Ibadan is classified as oxic paleustalf, Alagba soil series (Greenland, 1981) while in Jos plateau the soil is Ferruginous tropical soils (Kowal and Knabe, 1972).

Data collection and analyses

In the growth chamber and field studies, diffusive resistances of the lower (abaxial) leaf surface were measured at 3, 6, 9 and 12 months after planting from 3 fully-expanded leaves. Measurements were done with an L1-1600 steady state porometer (L1-Cor 1600, L1-Cor Inc., USA) from 900 to 1500 h. The total dry biomass was determined by harvesting the central 4 plants in each plot at the end of the growing cycle (12 months after planting) and data obtained from Jos plateau was used to calculate correlations with stomatal conductance.

Statistical analytical system (SAS, 1996) programme was used for data analyses. Analysis of variance and Duncan's multiple range test were used to evaluate significant differences in data collected.

RESULTS

Growth chamber study

The total dry biomass was significantly reduced at low temperatures (25/20°C and 20/15°C) than at ambient (32/22°C) at 12 MAP (Table 1). In assessing the response of stomatal conductance to temperature regimes at midday, measurements were taken in the morning and afternoon and the average values were used in this report. The ambient temperature had highest values whereas the temperature regime of 20/15°C recorded the lowest values throughout the growing period (Table 1). The leaf conductance increased as the temperature increased. Thus, cassava plants subjected to regime 20/15°C had the lowest leaf conductance values of 0.3, 0.35, 0.32 and 0.36 mol m⁻²s⁻¹ at 3, 6, 9 and 12 MAP respectively (Table 1).

Genotypic variations were observed for total dry matter throughout the sampling periods (Table 2). TMS 30572 and TME1 produced the highest dry biomass per plant at low temperatures when compared to other genotypes tested (Table 2). Also, genotypic differences in leaf stomatal conductance among the temperature regimes were recorded during the sampling periods (Table 3). TMS 30572, Danwaru, TMS 91934 and TME1 had the highest values of stomatal conductance throughout the sampling periods whereas the lowest values were recorded for TMS 4(2)1425 and Danduala (Tables 3 and 4).

Field study

The total dry biomass was significantly reduced at Jos plateau than at Ibadan at 12 MAP (Table 1). At Ibadan, TMS 30572 and TMS 91934 performed best whereas at

Table 1. Environmental effects on stomatal conductances and total dry matter of 8 cassava genotypes during 1994 and 1995 crop seasons.

Field study	Months after planting (map)			
	3 map	6 map	9 map	12 map
	stomatal conductances (mol m⁻²s⁻¹)			
Ibadan, 1994/1995	11.0 ^a	21.0 ^a	28.4 ^a	18.5 ^a
Ibadan, 1995/1996	10.4 ^a	19.3 ^a	20.5 ^a	17.3 ^a
Jos, 1994/1995	6.7 ^b	6.5 ^b	5.7 ^b	6.8 ^b
Jos, 1995/1996	5.6 ^b	6.8 ^b	6.8 ^b	7.2 ^b
	Total dry matter (t/ha)			
Ibadan, 1994/1995	2.4 ^a	6.9 ^a	7.2 ^a	11.2 ^a
Ibadan, 1995/1996	2.2 ^b	3.2 ^b	4.2 ^b	5.1 ^b
Jos, 1994/1995	1.7 ^c	2.4 ^c	3.2 ^c	3.9 ^c
Jos, 1995/1996	1.3 ^d	1.8 ^d	2.9 ^d	2.3 ^d
Growth chamber study	stomatal conductances (mol m⁻²s⁻¹)			
32/22 °C	0.40 ^a	0.55 ^a	0.63 ^a	1.12 ^a
25/20 °C	0.25 ^b	0.42 ^b	0.41 ^b	0.60 ^b
20/15 °C	0.13 ^c	0.35 ^c	0.32 ^c	0.36 ^c
	total dry matter (g) / plant			
32/22 °C	70 ^a	200 ^a	250 ^a	400 ^a
25/20 °C	70 ^a	150 ^b	241 ^b	350 ^b
20/15 °C	70 ^a	112 ^c	160 ^c	300 ^c

Means in the same vertical column with different superscripts and within the same study and also in the same growth parameter are significantly different ($p < 0.05$).

Table 2. Total dry biomass of eight cassava genotypes as affected by temperature regimes (32/22 °C, 25/20 °C and 20/15 °C) in growth chambers and field conditions (Ibadan and Jos plateau) during 1994 and 1995 crop seasons.

Genotype	Growth chamber study			Field study	
	Dry biomass per plant (g)			Dry biomass (t/ha)	
	32/22 °C	25/20 °C	20/15 °C	Ibadan	Jos plateau
Danduala	152 ^b	114 ^b	83 ^a	6 ^c	2 ^c
TMS 30555	185 ^{ab}	141 ^a	95 ^a	16 ^{ab}	3 ^{bc}
TME1	200 ^a	126 ^b	83 ^a	13 ^{ab}	10 ^a
TMS 4(2)1425	165 ^b	119 ^b	95 ^a	15 ^{ab}	2 ^c
Danwaru	137 ^b	106 ^b	83	5 ^c	5 ^b
TMS 91934	162 ^b	120 ^b	92 ^a	17 ^{ab}	4 ^b
TMS 30001	184 ^{ab}	144 ^a	91 ^a	14 ^b	3 ^{bc}
TMS 30572	209 ^a	156 ^a	94 ^a	22 ^a	7 ^a

Means in the same vertical column with different superscripts are significantly different ($P < 0.05$).

Jos plateau TMS 30572 and TME1 performed better than other genotypes (Table 2). The leaf stomatal conductances were significantly higher at Ibadan as compared to Jos plateau (Table 3). Genotypic differences in leaf stomatal conductances were observed across locations and years (Tables 3 and 4). At Jos plateau, TMS 91934, TMS 30572 and Danwaru had the highest values for leaf conductances (Table 3). The correlation of dry biomass obtained from cassava plants grown at Jos plateau with

leaf stomatal conductances showed significant differences ($P < 0.05$) in leaf stomatal conductances ($r = 0.77$, $n = 8$). Significant genotype temperature was observed on leaf stomatal conductances at 12 months after planting (Table 4).

DISCUSSION

The results of this study indicate that the ambient tempe-

Table 3. Effect of temperature x genotype on stomatal conductances across sampling periods in growth chamber study (20/15°C, 25/20°C) and field study (Jos plateau and Ibadan) during 1994 and 1995 crop seasons.

Genotype	Stomatal conductances (mol m ⁻² s ⁻¹)			
	20/15°C	25/20°C	Jos plateau (18 ± 5°C)	Ibadan (27 ± 6°C)
Danduala	0.2	0.5	3.95	12.2
Danwaru	0.5	0.7	6.05	15.2
TMS 4(2)1425	0.3	0.8	4.68	12.4
TMS 30001	0.4	0.5	5.34	11.9
TMS 30555	0.3	0.6	4.98	7.6
TMS 30572	0.6	0.8	6.82	19.9
TMS 91934	0.4	0.7	5.01	17.7
TME1	0.5	0.6	5.47	16.2

When the F-values were significant from the analysis of variance for interaction effects, least significant differences at P < 0.05 were calculated and it was 0.3

Table 4. Genotype effect on stomatal conductances at 12 months after planting in growth chamber study and field study during 1994 and 1995 crop seasons.

Genotype	Stomatal conductances (mol m ⁻² s ⁻¹)	
	Growth Chamber Study	Field Study
TMS 30572	0.6 ^a	11.04 ^a
TMS 30555	0.4 ^c	10.05 ^a
TME1	0.5 ^b	12.03 ^a
TMS 4(2)1425	0.4 ^c	9.05 ^b
TMS 30001	0.4 ^c	8.75 ^b
Danwaru	0.5 ^b	12.05 ^a
TMS 91934	0.6 ^a	10.50 ^a
Danduala	0.7 ^a	7.75 ^b

Means in the same vertical column with different superscripts are significantly different (P < 0.05).

ture (32/22°C) and Ibadan had the highest stomatal conductances on the abaxial leaf surfaces than low temperatures (25/20°C and 20/15°C) and Jos plateau. These observations imply that the stomata in cassava leaves reacted directly to changes in temperatures and closed rapidly in low temperatures, thus restricting CO₂ supply and causing a decline in photosynthesis. Similar results have been reported by El-sharkawy and Cock, (1990) who observed that in cassava plants the leaf stomatal conductances increase with rising temperature up to an optimum between 25 and 35°C. Also, it has been reported that leaf conductances in plants decreases when plants are subjected to low temperature (Kramer, 1983). The decrease in leaf conductances is a result of stomatal closure and a decrease in root hydraulic conductivity (Running and Read, 1980). The reduced leaf conductances observed among cassava plants grown

under low temperature regimes imply stomatal closure, which inhibits photosynthetic gas exchange levels. This response may partially explain, why cassava is rarely grown in regions with mean temperature less than 15°C.

There were genotypic differences in leaf stomatal conductances among the temperature regimes and locations. Danwaru TMS 30572, TMS 91934 and TME1 had higher leaf conductances than the other genotypes while TME1 and TMS 91934 had higher leaf stomatal conductance than other genotypes. This implies that stomata response to leaf conductance is most sensitive and it is affected by genetical and environmental factors. Also, at low temperatures, TMS 91934, TMS 30572, Danwaru and TME1 seem to have higher leaf stomatal conductances than other genotypes but genotypic differences were inconsistent across the sampling dates. The results revealed these genotypes seem to be less affected by low temperature stress when leaf stomatal conductance was used as an indicator.

This study showed significant G x E interaction for leaf stomatal conductances which will be useful to identify stable genotype across environments especially the mid altitudes. These results agree with observations by Manrique (1990), Akparobi et al. (2002) who reported genotypic differences for adaptation to high and low temperature conditions.

Conclusions

Deduction from this study showed that high stomatal conductances are interpreted to mean greater carbon assimilation rate in cassava. The higher value of stomatal conductance for Danwaru, TMS 91934, TMS 30572 and TME1 at Jos plateau may be responsible for better dry biomass yield. The correlation of dry biomass with leaf stomatal conductance suggest that high stomatal conductance are important physiological characteristics of cas-

sava genotypes tolerant to low temperatures.

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