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Identification of Peanut Genotypes with High Water Use Efficiency under Drought Stress Conditions from Peanut Germplasm of Diverse Origins

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Abstract: The aims of this study were to investigate the effect of drought stress on Total Dry Matter (TDM), pod yield, Water Use Efficiency (WUE), harvest index (HI), SPAD Chlorophyll Meter Readings (SCMR), Specific Leaf Area (SLA) and canopy temperature, to identify drought resistant peanut genotypes from a collection of peanut germplasm and to establish the relationships among drought resistance traits. Field experiments was conducted in a strip plot design with four water regimes (field capacity (FC), 25, 40 and 60 reduction percentage of amount of water regimes in FC) as main and sixty peanut genotypes as sub-treatments. Observations on TDM, pod yield and SLA were measured at harvest. SCMR and canopy temperature were recorded at 30, 60 and 90 day after emergence. WUE were computed using the data on amount of water input and TDM. HI was computed using the data on pod yield and TDM. The result showed that the effects of drought reduced TDM, pod dry weight, HI, WUE and SLA, but increased SCMR and canopy temperature. The correlation of WUE was positively related to SCMR under water limit conditions. The surrogate traits with well associated on WUE could be useful as selection criteria for drought tolerance. In this germplasm, the identical genotypes with high WUE in all of drought levels were Tifton-8, 14 PI 430238 and 205 PI 442925. KK 60-3, 101 PI 268659 only found high WUE in severe drought condition. The genotypes identified might be useful in future breeding programs for drought tolerance.

Key words: Water stress, SPAD chlorophyll meter reading, canopy temperature, specific leaf area and drought tolerance

INTRODUCTION

Peanut is grown widely under rain-fed conditions in the semi-arid tropics, where, drought is a major constrain of peanut productivity especially during the pod and seed forming stages that can greatly reduce pod yield (Songsri *et al*., 2008a, b). Drought resistant varieties have been used to stabilize peanut productivity under drought conditions. Breeding for drought resistance has been an important strategy in alleviating the problem.

The identification of drought resistance germplasm is an important stage of breeding for drought resistance. However, large collections of peanut germplasm lines have been rarely screened for drought resistance and the studies conducted so far have been limited to small numbers of peanut genotypes because of the difficulty of screening procedures. Yield has been a primary target trait of drought resistance breeding in peanut and selection for yield has slow progress because of the complex nature of the trait that causes high genotype by environment interactions (Branch and Hildebrand, 1989; Jackson *et al*., 1996; Araus *et al*., 2002). Therefore, alternative selection strategies in order to breed for drought resistance are worth exploring. Water Use Efficiency (WUE) or Transpiration Efficiency (TE) is one of such traits that can contribute to productivity when water resources are limited (Wright *et al*., 1994). WUE might be suitable for use as a selection criterion for drought resistance and it is used to express the amount of total biomass produced/unit of water use in evapotranspiration (Teare *et al*., 1982). WUE is based on the total dry matter produced and total water used at the end of the season and there are significant differences between genotypes (Matthews *et al*., 1988). The genotypic variation for yield, transpiration, water use efficiency and harvest index has been demonstrated in both greenhouse and field conditions (Matthews *et al*., 1988; Hubick *et al*., 1986; Wright *et al*., 1994; Nageswara *et al*., 1988).

In general, high yield is correlated with high WUE (Kramer, 1983). Peanut genotypes with high WUE under drought conditions are considered to be drought tolerant in terms of total dry matter production (Nautiyal *et al*., 2002). However, the selection through this process is difficult or even unsuccessful due to genotypes and environmental variations (Arunyanark *et al*., 2008). This hinders the progress in breeding for drought resistance. The identification and use of surrogate traits for WUE

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that are simple and have low environmental variations under drought conditions would be more effective and efficient.

The relationships on WUE related physiological traits demonstrated in peanut, there were close relationships between SPAD Chlorophyll Meter Reading (SCMR) and WUE. Chlorophyll content can be measured simply and rapidly by handheld portable SPAD chlorophyll meter and the association between chlorophyll content and SPAD reading was high and positive (Samdur *et al*., 2000; Arunyanark *et al*., 2008). Sheshshayee *et al*. (2006) investigated the relationship between SCMR and WUE in six peanut genotypes with wide genetic variation for SLA. The study was conducted two pot experiments in dry and rainy seasons. They found that there was a significant positive relationship between SCMR and WUE and negative relationship between SLA and WUE. Wright *et al*. (1994) also found negative relationship between SLA and WUE in four peanut genotypes under two water regimes. Rucker *et al*. (1994) found that peanut canopy temperatures were correlated with visual drought stress rating and yield. In plant breeding program, the interest is in finding genotypes that maintain low canopy temperature under field conditions. However, more previous studies investigated in a few peanut genotypes and water regimes. The investigation in a large number of peanut genotypes and different levels of soil moisture gradients will provide useful information for explaining the relationships between surrogate traits and WUE.

In this study, the effects of drought stress on Total Dry Matter (TDM), pod dry weight, WUE, HI, SCMR, SLA and canopy temperature were reported. Drought resistant peanut genotypes from a collection of peanut germplasm were identified based on yield and other surrogate traits of drought resistance and the relationships among drought resistance traits were established.

MATERIALS AND METHODS

The experiment was conducted under field conditions in the dry season during November 2005 to March 2006 at the Field Crop Research Station of Khon Kaen University located in Khon Kaen province, Thailand (latitude 16°28′ N, longitude 102°48′ E, 200 m above mean sea level). Soil type is Yasothon series (Yt: fine-loamy; siliceous, isohypothermic, Oxic Paleustults). For each plot, there were two rows with 3.2 m in length with spacing of 50 cm between rows and 20 cm between hills in a row.

A strip-plot design with four replications was used. Four water treatments were assigned as factor A and 60 peanut germplasm lines of diverse origins were assigned as factor B. A line-source sprinkler system (Hank *et al*., 1976) was installed at the center of the experimental field

Table 1: Sixty peanut genotypes diverse in source countries

No. of entries	Identification	Source countries	No. of entries	Identification	Source countries
1	248 Grif 13911	China	31	12 PI 430233	China
2	265 Grif 13256	China	32	45 PI 430234	China
3	35 Grif 13932	China	33	305 PI 430236	China
4	269 PI 157542	China, Jiangxi	34	306 PI 430237	China, Liaoning
5	88 PI 157547	China, Jiangxi	35	14 PI 430238	China
6	89 PI 157549	China, Jiangxi	36	15 PI 433347	China
7	90 PI 157551	China, Jiangxi	37	45 PI 433348	China
8	37 PI 158838	China, Jiangxi	38	187 PI 433352	China
9	97 PI 158854	China, Jiangxi	39	190 PI 433356	China
10	100 PI 162604	China	40	194 PI 436545	China
11	283 PI 234375	China	41	196 PI 436547	China
12	101 PI 268659	China	42	197 PI 436548	China
13	102 PI 268660	China	43	198 PI 436549	China
14	285 PI 268832	China	44	200 PI 442566	China, shandong
15	104 PI 268884	China	45	204 PI 442572	China, shandong
16	287 PI 268885	China	46	205 PI 442925	China, guandong
17	105 PI 268888	China	47	ICGV 98300	ICRISAT[1]
18	106 PI 268949	China	48	ICGV 98303	ICRISAT[1]
19	289 PI 268950	China	49	ICGV 98305	ICRISAT[1]
20	290 PI 269060	China	50	ICGV 98308	ICRISAT[1]
21	111 PI 291251	China	51	ICGV 98324	ICRISAT[1]
22	295 PI 295754	China	52	ICGV 98330	ICRISAT[1]
23	3 PI 313157	China	53	ICGV 98348	ICRISAT[1]
24	43 PI 313160	China	54	ICGV 98353	ICRISAT[1]
25	5 PI 313160	China	55	Tainan 9	KKFCRC[2] (Thailand)
26	299 PI 313163	China	56	KK 60-3	KKFCRC[2] and KKU[3] (Thailand)
27	301 PI 430226	China	57	Tifton-8	USDA[4]
28	9 PI 430227	China	58	Non-nod	ICRISAT[1]
29	303 PI 430230	China	59	KKU 60	KKU[3] (Thailand)
30	11 PI 430231	China	60	(Luhua 11×China 97-2) F6-8-2	Thailand

ICRISAT 1: International Crop Research Institute for the Semi-Arid Tropics, KKFC 2: Khon Kean Field Crop Research Centre, KKU 3: Khon Kean University and USDA 4: United State Department of Agriculture

to supply water to the crop at four water gradients designated as, field capacity (100%); FC, 75 of FC, 60 of FC and 40% of FC. The four water gradients were hereafter referred to as W1, W2, W3 and W4, respectively and they were placed horizontally along the line source sprinkler at the distances from the center of 1-4, 4-7, 7-10 and 10-13 m, respectively. Water content of each level was measured by catch cans (24 cans for each water regime treatment). Soil moisture content was also monitored weekly by neutron probe at the depths of 30, 60 and 90 cm form soil surface (6 tube for each water regime treatment). The list of peanut genotypes used in this study is provided in Table 1.

Crop management: Disc plowing was performed three times to prepare soil suitable for the experiment. Lime ($CaCO_3$) at the rate of 625 kg ha^{-1} was incorporated into the soil during soil preparation. Phosphorus fertilizer as triple superphosphate at the rate of 122.3 kg ha^{-1} and potassium fertilizer as potassium chloride at the rate of 62.5 kg ha^{-1} were applied shortly prior to planting. Seeds were treated with captan (3a, 4, 7, 7a-tetrahydro-2-[(trichloromethyl)thio]-1H-isoindole-1, 3(2H)-dione) at the rate of 5 g kg $^{-1}$ seed before planting. The seeds were over-planted and the seedlings were thinned to one plant/hill at 7 days after emergence (DAE). Gypsum ($CaSO_4$) at the rate of 312 kg ha^{-1} was incorporated into the soil at 15 days after emergence (DAE). Carbofuran (2, 3-dihydro-2, 2-dimethylbenzofuran-7-ylmethylcarbamate 3% granular) was applied at the pod setting stage. Pest and diseases were controlled by weekly applications of carbofuran [2, 3-dihydro-2, 2-dimethylbenzofuran-7-ylmethylcarbamate 3% granular (dibutylaminothio) methylbamate 20% w v $^{-1}$, water soluble concentrate] at 2.5 L ha^{-1}, methomyl [*S*-methyl-*N*((methylcarbamoyl) oxy thioacetimidate 40% soluble powder] at 1.0 kg ha^{-1} and carboxin [5,6-dihydro-2-methyl-1, 4-oxath-ine-3-carboxanilide 75% wettable powder] at 1.68 kg ha^{-1}.

Irrigation: Prior to planting, water was supplied uniformly to the experimental field at water holding FC at the depth of 30 cm using minisprinkler to facilitate uniform emergence and crop establishment until 15 DAE. Different water gradients were supplied by the line source sprinkler system to the crop at 15 DAE until harvest. W1 was used as a control treatment and maintained at FC until harvest. The rest of water treatments (W2-W4) are proportional to soil moisture content at FC with reducing soil moisture contents, while the distances were increasing to marginal fields as described previously (Fig. 1). As water supplied to the crop was controlled for FC only and the rest treatments were proportional to the control automatically, water was added to the experiment based on crop water requirement and surface evaporation which were calculated following the methods described by Songsri *et al.* (2008a).

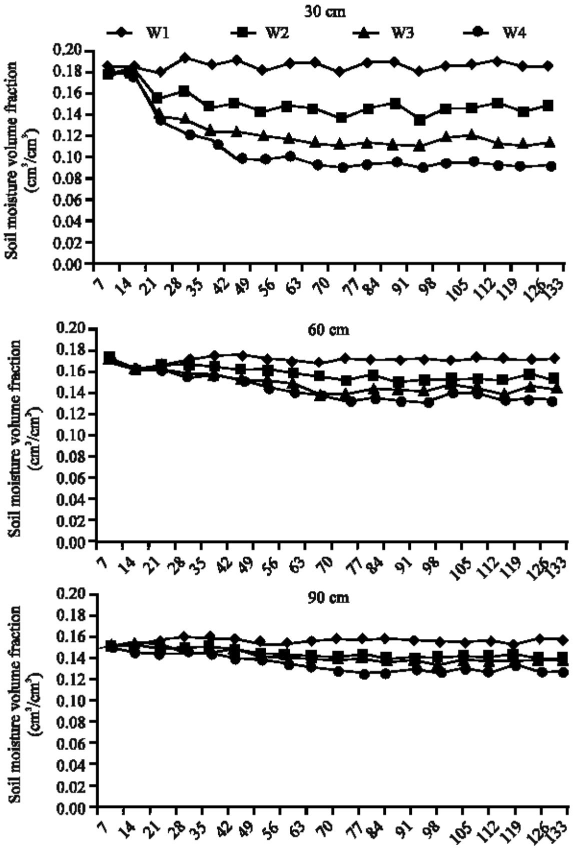

Fig. 1: Soil water content of three soil depth (30, 60 and 90 cm from soil profile) in the 4 water treatments (W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction % of amount of water regimes in FC, respectively)

The calculation of total crop water use for each water treatment was calculated as the sum of transpiration and soil evaporation. Transpiration (T) was calculated using the equation:

$$Etcrop = ET_0 \times K_c$$

Where:

Etcrop	=	Crop water requirement (mm day^{-1})
Et_0	=	Evapotranspiration of a reference plant under specified conditions calculated by pan evaporation method
K_c	=	The crop water requirement coefficient for peanut, which varied depending on growth stages. Surface evaporation was calculated as:

$$E_s = \beta \times (E_0/t),$$

Where:

E_s = Soil evaporation (mm)

β = Light transmission coefficient measured depending on crop cover

E_0 = Evaporation from class A pan (mm day^{-1})

t = Days from the last irrigation or rain (days)

Data collections

Soil moisture content and meteorological conditions: Soil moisture were measured at planting and harvest at the depths of 30, 60 and 90 cm from soil surface using micro augur method to check whether the water treatments were under control. Soil moisture contents were also detected by neutron probe at weekly intervals throughout the course of the experiment to monitor water that supplied to the crop if it was correct amount. Rainfall, Relative Humidity (RH), evaporation (E_0), maximum and minimum temperature and solar radiation were recorded daily from sowing until harvest by a weather station located 100 m away from the experimental field.

SCMR, canopy temperature and SLA: A Minolta SPAD-502 m (Tokyo, Japan) was used to record SCMR at 30, 60 and 90 DAE on the four leaflets from each leaf as described by Nageswara *et al*. (2001). SCMR was measured during 9.00-11.00 am, using the second fully expanded leaf from the top of main stem, totally 5 leaves for each plot and, then, single value was obtained for each plot by averaging the data.

Canopy temperature was measured from 3 plants for each plot at 12.00-14.00 am at 30, 60 and 90 DAE using an infrared thermometer (Testo 830-T1, Testo Inc., Germany).

SLA was measured at harvest by taking 50 leaves from 16 plants randomly selected from each plot. Leaf area was measured using a leaf area meter (ACC-400, Hayashi Denken, Japan) and the leaf samples were oven dried at temperature 80°C at least 48 h to determine the leaf dry weight. Then, SLA was calculated using the relationship as follows:

$$SLA = \text{leaf area (cm}^2\text{)/leaf dry weight (g)}$$

Water use efficiency (WUE) calculation: Evapotranspiration (ET) under varying water regimes was calculated using the soil water balance equation for the growing season as follows:

$$ET = I+(Mi-Mf)-D-R$$

Where:

I = The irrigation applications

M_i = Starting soil moisture before sowing

M_f = Soil moisture at final harvest (soil moisture was measured by gravimetric method)

D = Soil water drainage

R = Surface runoff

Percolation and surface runoff were assumed to be not significant and the values were ignored. WUE was estimated using the formula proposed by Teare *et al*. (1982):

$$WUE = \text{Dry matter yield/water used in evapotranspiration}$$

Total dry weight (TDM), pod yield and HI: At, harvest, the plants at two ends of the rows were discarded. As plants were bordered by the adjacent plots, all plants in an area of 3.2 m^2 were harvested without discarding the border rows. The plants were cut at soil surface and depodded in the field. Fresh shoot weight was measured in the field. A random shoot sample of 2 kg was taken, weighed, partly dehydrated by expose to the sun and then oven-dried at 80°C for 48 h until constant weight. The dry sample was weighted and the dry weight of the sample was converted to dry weight of the plot. Pods were air-dried and weighted then TDM was calculated using shoot dry weight and pod weight, excluding root weight. HI was calculated from pod dry weight divide by total biomass.

Statistical analysis: Analysis of variance was performed for each character followed a strip plot design (Gomez and Gomez, 1984). When the differences of main effects were significant (p = 0.05), Dancan's multiple rang test was used to compare means. Simple correlation coefficients among pod dry weight, TDM, HI and WUE and SCMR, SLA, canopy temperature and WUE were calculated for all water levels to determine the relationships among characters under investigation under different water conditions. All calculations were performed using MSTAT-C package.

For the traits with multiple date evaluation such as SCMR and canopy temperature, the most appropriate evaluation times were selected because of high F-ratios and low CV values from analysis of variance. The coefficients of variation for SCMR at 30, 60 and 90 DAE were 6.53, 7.17 and 10.57, respectively, whereas the F-ratios were 6.36, 8.39 and 7.68, respectively. Therefore, 60 DAE was selected. The appropriate evaluation time for canopy temperature was selected by the same criteria. The coefficients of variation for canopy temperature at 30, 60 and 90 DAE were 1.54, 0.95 and 2.33, respectively and the F-ratios were 6.56, 5.72 and 4.84, respectively. Therefore, 90 DAE was selected and reported.

RESULTS AND DISCUSSION

Identification of germplasm suitable for use in breeding programs is always important for the success in developing certain characters. In this study, we identified some promising germplasm lines of peanut with good performance for total dry matter, pod yield, water use efficiency and its surrogate traits. We also, reported that drought stress at different levels of severity had different effects on these characters and the relationships between water use efficiency and traits related to pod yield and water use efficiency and its surrogate traits (SCMR, SLA and canopy temperature) were identified.

Soil moisture content and meteorological conditions: Figure 1 showed soil moisture contents of different water regimes at three depths (30, 60 and 90 cm) of soil profile across the experiment. Soil moisture contents of different water regimes (W1-W4) were clearly separated at the soil depth of 30 cm, starting from 22 DAE when drought was imposed to the crop by line-source sprinkler irrigation system for a week. The differences in soil moisture content among water regimes were reduced with the depth of the soil profile. The results showed adequate control of water treatments.

As can be seen in Figure 2, the field was planted on 11 November 2005 and harvested on 10-23 March 2006 and there was a 13 mm rainfall at 100 days after planting (DAP). The excess soil moisture because of rainfall would not have significant effect on the crop because it was small amount, the soil is highly sandy and it occurred at the late growth stage near harvest. Mean air temperatures ranged from 18.8 to 32.3°C during crop season.

The effect of drought to yield and drought tolerance traits: Analysis of variance showed significant differences among water regimes for pod yield, TDM, HI, WUE, specific leaf area (SLA), SCMR and canopy temperature (Table 2). However, the differences among peanut genotypes were significant for most characters investigated except for specific leaf area. The low variation in specific leaf area could be due to the fact that the evaluation at harvest was too late. The suitable times fore assessing SLA in peanut would be about 60 DAE (Nigam and Aruna, 2007). The interactions between water regimes x genotypes were significant for pod yield, TDM, HI and WUE, but not significant for SLA, SCMR and canopy temperature (Table 2). Previous studies reported both supporting and contrasting results. The interactions between genotypes and water regimes were larger for TDM and WUE than for chlorophyll content, which is closely related to SCMR (Arunyanark *et al.*, 2008). However, Wright *et al.* (1994) found that genotype×water treatment interaction was not significant for WUE. This could be due to the differences in materials used and the differences in experiment conditions. In general, SCMR, canopy temperature and SLA had lower G×E interactions than did pod yield, TDM, WUE and HI because they are more complex traits (Branch and Hildebrand, 1989; Jackson *et al.*, 1996; Araus *et al.*, 2002). Because of low

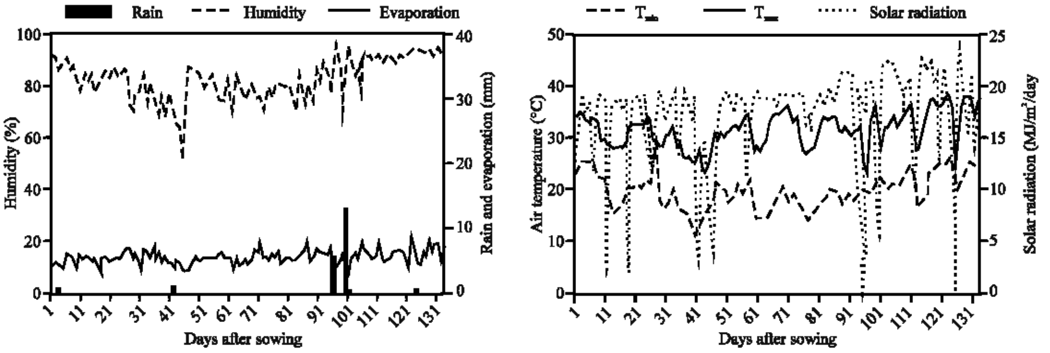

Fig. 2: The meteorological conditions during season (2005/06) (a: Rain fall, humidity and evaporation; b: maximum temperature (T_{max}), minimum temperature (T_{min}) and solar radiation)

Table 2: Mean square of Total Dry Matter (TDM), pod yield, Harvest Index (HI), Water Use Efficiency (WUE) and Specific Leaf area (SLA) at harvest; SPAD Chlorophyll Meter Reading (SCMR) and Canopy Temperature (CT) at 30, 60 and 90 day after emergence (DAE)

							SCMR			CT		
Source	df	TDM	Pod yield	HI	WUE	SLA	30 DAE	60 DAE	90 DAE	30 DAE	60 DAE	90 DAE
Replication	3	11420000	2722685	0.0952	1.771	17972	104.5	33.5	432.3	1.64.8	151.7	2031.5
Water	3	342000000**	23140000**	0.4276**	2.614**	57350*	1196.6**	3796.7**	8218.7**	794.1 ns	1507.2**	1238.3**
Error (a)	9	1401056	477123	0.0024	0.13	10465	16.85	21	70.2	209	22.3	41
Genotypes	59	5113055**	360342**	0.0191**	1.103**	682 ns	54.5**	92.5**	161**	4.5**	2 ns	5.3**
Error (b)	177	716705	36157	0.0012	0.142	903	8.6	11	20.9	2.9	2.1	2.2
Water×genotypes	177	588643**	74298.8**	0.0018**	0.223**	757 ns	3.8ns	8.9 ns	12.5 ns	2.2 ns	2.1 ns	1.7 ns
Error (c)	531	312459	27394.7	0.0009	0.063	761	3.2	8	10.3	1.8	1.7	1.6

**, *: Significant at 1, 5% level and ns: Not significant

Table 3: Means and reduction percentage on total dry matter (TDM), pod yield, harvest index (HI), water use efficiency (WUE) and specific leaf area (SLA) at harvest; SPAD chlorophyll meter reading (SCMR) at 60 day after emergence (DAE) and canopy temperature (CT) at 90 DAE

Treatment	TDM (reduction %) ------------ ($kg\ ha^{-1}$)	Pod yield (reduction %) ------------ ($kg\ ha^{-1}$)	HI (reduction %)	WUE (reduction %) --------- ($g\ L^{-1}$)	SLA (reduction %) ------------ ($cm^2\ g^{-1}$)	SCMR (reduction %) ----------- 60 DAE	CT (reduction %) --------------- (°C) 90 DAE
Water regimes							
W1	5324.3a	867a	0.161a	1.848a	168a	41.62d	23.9c
W2	4606.6b (13)	624b (28)	0.135b (16)	1.697b (9)	153ab (8)	44.82c (-8)	24.1c (-1)
W3	3573.1c (33)	326c (62)	0.093c (42)	1.657b (11)	140b (16)	48.1b (-16)	25.8b (-8)
W4	2595.1d (51)	170d (80)	0.066d (59)	1.6472b (12)	132b (21)	50.79a (-22)	29.3a (-23)

Different letters adjacent to data in the same column show significance at $p<0.05$ by Duncan's multiple range test, W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction% of amount of water regimes in FC, respectively, Values in parenthesis are reduction percentage with are calculated by [100- ((the data of stress treatment <W2, W3 or W4>×100)/The data of non-stress treatment)]

G×E interactions, the use of surrogate traits as selection criteria might be useful for improving WUE if heritabilities are high. High heritability estimates for SCMR and SLA have been reported (Upadhyaya, 2005). Therefore, they are promising as selection criteria for WUE.

Drought stress significantly reduced TDM, pod dry weight, HI, WUE and SLA, but it increased SCMR and canopy temperature (Table 3). The more severe drought stresses the more reductions in TDM, pod yield, HI and SLA. Our results supported those of previous studies (Nageswara Rao *et al*., 1985; Arunyanark *et al*., 2008). The reduction in WUE was smallest when compared with those of TDM, pod yield, HI and SLA. The reductions in TDM were 13, 33 and 51% for W2, W3 and W4, respectively and the reductions in pod yield were 28, 62 and 80%, respectively (Table 3). Similar to those for TDM and pod yield, the reductions in HI accounted for 16, 42 and 59, respectively (Table 3). However, the reductions in WUE were minimal, accounting for only 9, 11 and 12%, respectively and the differences among water regimes was found only between stressed treatment and non-stressed treatment only. SCMR and canopy temperature increased sharply when subjected to severe drought stress. The increases in SCMR were 8, 16 and 22% for W2, W3 and W4, respectively and the increases in canopy temperature were 1, 8 and 23%, respectively (Table 3).

The sensitivities in response to drought stress for pod yield, TDM and HI were higher than that for WUE. Previous studies found that the reductions in pod yield and TDM were 68-80% and 37-60% compare with well-water conditions, respectively (Nageswara Rao *et al*., 1985). In this study, drought stress did reduce WUE of 9-12%, but there were certain genotypes showing positive response under drought conditions (Table 4). In contrast to this study, Arunyanark *et al*. (2008) found general increase in WUE in response to drought stress. However, they also found the reduction in WUE in certain peanut genotypes. Hubick *et al*. (1988) found that WUE varied significantly among genotypes irrespective of whether peanuts were drought-stress or well-watered. Drought stress also reduces other characters such as nitrogen fixation and related traits (Venkateswarlu *et al*., 1990; Pimratch *et al*., 2008).

Drought is known to affect chlorophyll content in many crops including, wheat (Sarker *et al*., 1999), grass *Eragrostis curvula* (Colom and Vazzana, 2003), cattail (*Typha latifolia*) (Li *et al*., 2004) and turfgrasses (Jiang and Huang, 2001) thereby inhibiting photosynthetic capacity (Epron and Dreyer, 1993). The ability to maintain chlorophyll density under water deficit conditions has been suggested as a drought resistance mechanism in peanut (Arunyanark *et al*., 2008; Sheshshayee *et al*., 2006). In present study, SCMR was increased by water limit conditions. The increase in SCMR, the trait related to photosynthetic capacity, could contribute to drought tolerance.

The increase in canopy temperature was not unexpected. There were the higher canopy temperature under drought stress conditions than under well-irrigated conditions in wheat (Siddique *et al*., 2000) and rice (Dennis and O'Toole, 1995). The peanut genotypes with lower canopy temperature are preferable because they have higher transpiration and, therefore, have higher carbondioxide exchange rate than the genotypes with high canopy temperature.

Interaction between genotypes and water regimes led us to separate analysis for each water regime. However, the interactive variances were much smaller than variances of genotype main effect and means across water regimes were presented. The identification of peanut genotypes for each character was based on analysis of individual water regime instead of combined data (data not presented).

The peanut genotypes performed well for each character were identified. For total biomass production, Top-five genotypes under well-watered conditions were 14 PI 430238, 205 PI 442925, 12 PI 430233, Tifton-8 and KK 60-3. Tifton-8 and 14 PI 430238 had consistently high biomass production across four water regimes, whereas 205 PI 442925 had consistently high biomass production under the first three water regimes but not under the most severe drought (Table 4). Similarly, 12 PI 430233 had consistently high biomass production under well-water and mild drought only and the biomass production of this genotype reduced sharply under more severe drought. It is also, interesting to note here that KK 60-3 and 101 PI

Table 4: Means for total dry matter (TDM), pod yield, harvest index (HI) and water use efficiency (WUE) in 4 water treatment of selected peanut genotypes

	TDM (kg ha^{-1})				Pod yield (kg ha^{-1})			
Genotypes	W1	W2	W3	W4	W1	W2	W3	W4
Tifton-8	6306bc	6010ab	5443a	4021ab	466tuv	315uvw	179n-s	64o-r
14 PI 430238	8220a	6727a	5159abc	4175a	1570ab	1008abc	379d-l	182e-o
205 PI 442925	7981a	6378a	5389ab	3082c-h	1344b-f	900bcd	368d-l	119h-r
12 PI 430233	6584b	6437a	3418g-o	2526f-s	965f-o	743c-l	246i-s	140f-r
KK 60-3	6157bcd	5234b-f	4519a-e	3460abc	855g-t	818c-h	508a-d	372a
101 PI 268659	5831b-g	4942c-m	4660a-d	3923ab	1186b-g	824c-h	473a-f	341abc
KKU 60	5737b-h	5014c-k	3904d-k	2772c-o	1866a	1189a	483a-e	356ab
(Luhua 11×China 97-2) F6-8-2	5557c-n	4950c-l	3846d-k	2487f-t	1470bc	872b-f	466a-f	234c-i
11 PI 430231	4726j-s	4345g-s	2320p	2003p-u	1427bcd	1135ab	403b-i	276a-e
200 PI 442566	5154e-q	4313g-s	3522f-o	2283j-u	1391b-e	882b-f	454a-g	189e-n
198 PI 436549	4864g-r	4137l-s	3380g-o	2005p-u	1017e-m	749c-k	617a	247b-g
194 PI 436545	4679k-s	4337g-s	2815nop	1815stu	767h-u	710d-o	344d-o	237c-h
ICGV 98330	4942e-r	4374f-s	4051d-i	2929c-j	709i-v	507j-w	429b-h	241d-l
ICGV 98324	5185d-q	4507c-r	3675e-n	2789c-n	817g-u	476l-w	342e-p	165e-p
197 PI 436548	4929e-r	3954p-s	3972d-j	2396g-u	391v	332vw	263c-j	154f-r
89 PI 157549	3766s	3539s	3258h-p	2827c-m	681j-v	511i-w	366d-l	169e-p
289 PI 268950	6142bcd	4839c-o	4424b-f	3296b-e	861g-s	505j-w	295f-q	149f-r
106 PI 268949	5654bk	5028c-i	4038d-i	3160c-f	504s-v	354t-w	168o-s	95m-r
90 PI 157551	4519p-s	4173l-s	3465f-o	3029c-i	731h-u	598g-t	297f-p	156f-r
88 PI 157547	4273qrs	4139k-s	3821d-m	2696d-p	665l-v	530i-w	328d-p	163e-q

	HI				WUE (g L^{-1})			
Genotypes	W1	W2	W3	W4	W1	W2	W3	W4
Tifton-8	0.08st	0.05x	0.03v	0.02v	3.00ab	2.22ab	1.95bc	2.56ab
14 PI 430238	0.19c-i	0.15d-o	0.07j-v	0.04p-v	2.41c-g	2.48a	2.54a	2.65a
205 PI 442925	0.17c-o	0.14e-r	0.07m-v	0.04q-v	3.16a	2.35a	2.47a	1.96c-i
12 PI 430233	0.15d-q	0.11k-w	0.07m-v	0.05n-v	1.59k-s	2.32a	2.04b	1.61g-t
KK 60-3	0.14f-s	0.15c-m	0.11c-k	0.10b-h	2.87abc	1.93b-e	1.91bcd	2.20a-d
101 PI 268659	0.20b-e	0.17c-h	0.10e-o	0.09d-m	2.17d-j	1.82c-k	1.80b-g	2.35abc
KKU 60	0.32a	0.23ab	0.12cg	0.13abc	2.48c-f	1.85c-i	1.78b-h	1.76d-o
(Luhua 11×China 97-2) F6-8-2	0.27ab	0.18c-g	0.12c-f	0.09c-n	2.44c-g	1.82c-j	1.72c-n	1.58g-u
11 PI 430231	0.30a	0.26a	0.17ab	0.13ab	1.08t	1.60f-r	1.46j-s	1.27p-v
200 PI 442566	0.26ab	0.21bc	0.13cde	0.08d-o	2.24d-i	1.59f-r	1.60e-q	1.45k-v
198 PI 436549	0.21bc	0.18b-e	0.19a	0.12a-d	2.15d-j	1.52j-r	1.51g-r	1.27p-v
194 PI 436545	0.16c-o	0.16c-i	0.12c-g	0.14a	1.31rst	1.60f-r	1.45k-s	1.15tuv
ICGV 98330	0.14e-r	0.11i-v	0.10d-m	0.07h-r	2.58bcd	1.61e-q	1.53e-r	1.86d-k
ICGV 98324	0.16c-o	0.11l-x	0.09d-r	0.06j-t	2.34d-h	1.66c-p	1.60d-q	1.77d-o
197 PI 436548	0.06t	0.06wx	0.10e-o	0.06i-s	2.52b-e	1.46n-r	1.53e-r	1.52h-v
89 PI 157549	0.18c-l	0.14e-q	0.11c-k	0.06j-s	1.52l-t	1.30q-r	1.17s	1.80d-n
289 PI 268950	0.14g-s	0.10m-x	0.07l-v	0.05o-v	2.06e-k	1.78c-m	1.90bcd	2.10b-f
106 PI 268949	0.09q-t	0.07u-x	0.04s-v	0.03r-v	1.88h-n	1.85c-h	1.75b-k	2.01c-g
90 PI 157551	0.16c-o	0.14e-q	0.09e-r	0.06j-s	1.62ks	1.54h-r	1.40p-s	1.93c-j
88 PI 157547	0.15c-q	0.13f-t	0.09e-r	0.06i-s	1.78i-r	1.53j-r	1.32qrs	1.71e-p

Different letter adjacent to data in the same column show significance at $p<0.05$ by Duncan's multiple range test, W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction % of amount of water regimes in FC, respectively

268659 did not perform well under well-irrigated conditions but they performed well under severe drought (Table 4).

For pod yield (Table 4), the top-five genotypes under well-irrigated conditions were given to KKU 60, 14 PI 430238, (Luhua 11×China 97-2) F6-8-2, 11 PI 430231 and 200 PI 442566. The genotype with the most consistency for pod yield across four water regimes was KKU 60. This peanut genotype is a breeding line in advanced generation. 14 PI 430238 performed well only under well-irrigated conditions and mild drought conditions. KK 60-3, 198 PI 436549 and 101 PI 268659 showed high performance under severe drought but not under well-irrigated and mild drought conditions.

For HI, the top five genotypes under well-irrigated conditions were given to KKU 60, 11 PI 430231, (Luhua 11×China 97-2) F6-8-2, 200 PI 442566 and 198 PI 436549. 11 PI 430231, 198 PI 436549 and KKU 60 were the genotypes with the most consistency for HI across four water regimes. 200 PI 442566 performed well only under well-irrigated conditions and mild drought conditions (Table 4). 194 PI 436545 showed high performances under the most severe drought only.

For WUE, top five genotypes under well-watered conditions were 205 PI 442925, Tifton-8, KK 60-3, ICGV 98330 and 197 PI 436548. The genotype with the most consistency for WUE across four water regimes was Tifton-8, whereas 14 PI 430238 had consistently high WUE under the three stress treatments but not under non-stressed treatment (Table 4). 205 PI 442925 performed relatively well under normal and mild drought conditions but it performed poorly under the most severe conditions.

In contrast to 205 PI 442925, KK 60-3 and 101 PI 268659 performed well under the most severe drought, but performed poorly under normal and mild drought conditions (Table 4).

For SCMR (Table 5), top-five genotypes under well-watered conditions were (Luhua 11×China 97-2) F6-8-2, KKU 60, ICGV 98324, 14 PI 430238 and 89 PI 157549. (Luhua 11×China 97-2) F6-8-2, ICGV 98324, 14 PI 430238 and 205 PI 442925 were the genotypes with the most consistently high SCMR across four water regimes. KKU 60 performed well only under well-irrigated conditions. 289 PI 268950 showed high performance under the most severe drought, but not under first three water regimes.

Peanut genotypes with low canopy temperature are favorable. Top-five genotypes under well-watered conditions were 14 PI 430238, Tifton-8, 106 PI 268949, 205 PI 442925 and 90 PI 157551. The genotype with the most consistently low canopy temperature across four water regimes was 14 PI 430238. Tifton-8 had low canopy temperature only under well-irrigated conditions (Table 5). 88 PI 157547 showed rather low canopy temperature under the most severe drought, but not under first three water regimes (Table 5).

The relationships between yield and WUE, WUE and surrogate traits: Irrespective of water regimes, water use efficiency (WUE) was closely correlated with total dry matter (TDM) (Table 6). This could be due to the fact that WUE based on TDM produced and total water used (Matthews *et al*., 1988). Water use efficiency is important for biomass accumulation even under the most severe drought conditions and, therefore, it is an important criterion for drought tolerance.

In contrast to the previous relationship, the relationship between WUE and pod yield showed decreasing pattern with drought stress levels in positive direction, whereas the relationship between WUE and HI showed non-significant at non-stress and drought stress especially under the most severe drought stress conditions. For the relationship between WUE and pod yield, the correlation coefficient become non-significant at very severe drought stress.

Water use efficiency had high contribution to pod yield under well-watered conditions, but the contribution was reduced with severe drought stress. This could be due to low partitioning of biological yield to harvestable yield under drought stress conditions. The negative and

Table 5: Means for SPAD Chlorophyll Meter Reading (SCMR) at 60 Day After Emergence (DAE), Canopy Temperature (CT) at 90 DAE in 4 water treatment of selected peanut genotypes

	SCMR				Canopy temperature (°C)			
Genotypes	W1	W2	W3	W4	W1	W2	W3	W4
Tifton-8	42.7e-q	46.6c-j	48.6c-m	52.3a-k	26.8ij	30.4b-i	32.9a-g	33.4a-j
14 PI 430238	46.9a-c	50.4ab	54.1ab	53.1a-h	26.6j	29.0i	31.0jkl	32.3g-j
205 PI 442925	46.2b-e	52.5a	54.5ab	56.0ab	27.0g-j	30.5a-j	32.4a-k	33.8a-j
12 PI 430233	38.8q-m	40.2pq	45.1l-p	48.2i-q	28.6a-j	30.3c-i	31.9c-l	32.5f-j
KKU 60	48.8abc	48.2bcd	49.4c-j	52.8a-i	27.9b-j	29.4h-i	31.0jkl	32.6e-j
(Luhua 11×china 97-2) F6-8-2	50.3a	52.3a	52.1abc	6.7a	28.5a-j	31.1a-h	32.3b-k	33.9a-i
11 PI 430231	39.2p-y	44.3e-n	48.6c-m	48.0j-q	29.5abc	31.4a-g	33.3a-d	33.3b-j
200 PI 442566	41.9f-u	45.1d-n	49.4c-j	51.6b-o	28.8a-h	30.6a-i	32.8a-h	33.5a-j
198 PI 436549	41.5f-u	43.2i-q	49.1c-k	51.7b-n	28.8a-h	31.5a-g	33.4abc	34.6a-e
194 PI 436545	40.0l-y	43.7g-p	45.5j-p	49.7b-q	28.8a-h	31.3a-h	32.1b-l	32.4f-j
ICGV 98330	44.4c-j	49.4abc	51.9abc	51.8b-n	27.8c-j	32.3a-b	33.4abc	33.1b-j
ICGV 98324	47.4abc	47.7b-f	55.6ab	54.4ac	28.1b-j	30.8a-i	32.0c-l	33.6a-j
197 PI 436548	40.5i-y	43.6g-p	48.4c-m	53.3a-g	27.8c-j	30.8a-i	31.9c-l	33.6a-j
89 PI 157549	46.9a-d	48.5bcd	51.2bcd	54.1a-e	27.4e-j	30.4b-i	31.8d-l	33.0c-j
289 PI 298950	43.8c-m	45.3d-n	48.2c-m	55.2abc	27.9b-j	30.9a-i	30.6l	32.8e-j
106 PI 2689949	41.1h-x	45.8c-m	48.9c-l	51.8b-m	26.9hij	30.0e-i	31.8d-l	32.1hij
90 PI 157551	44.3c-j	45.7c-m	49.2c-k	52.1a-l	27.0g-j	29.8f-i	31.5f-l	34.1a-h
88 PI 157547	42.4e-s	45.2dn	47.6d-m	48.9f-q	27.5d-j	30.1di	31.5f-l	31.6j

Different letters adjacent to data in the same column show significance at p<0.05 by Duncan's multiple range test, W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction % of amount of water regimes in FC, respectively

Table 6: Relationships between water use efficiency (WUE) and total dry matter (TDM), pod yield, harvest index (HI) in 4 water treatment

	TDM				Pod yield				HI			
	W1	W2	W3	W4	W1	W2	W3	W4	W1	W2	W3	W4
WUE (W1)	1.000**				0.429**				0.0047 ns			
WUE (W2)		1.000**				0.357**				-0.0929 ns		
WUE (W3)			0.807**				0.340**				-0.0841 ns	
WUE (W4)				0.999**				0.132 ns				-0.3274**

**, *: Significant at 1 , 5 % level and ns: Not significant respectively, W1, W2, W3 and W4 in parenthesis are water regimes treatment (W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction% of amount of water regimes in FC, respectively)

Table 7: Relationships between water use efficiency (WUE) and SPAD chlorophyll meter reading (SCMR) at 60 day after emergence (DAE), canopy temperature (CT) at 90 DAE specific leaf area (SLA) at harvest in 4 water treatment

	SCMR				CT				SLA			
	W1	W2	W3	W4	W1	W2	W3	W4	W1	W2	W3	W4
WUE (W1)	0.144 ns				-0.355**				0.017 ns			
WUE (W2)		0.284**				-0.281*				0.220 ns		
WUE (W3)			0.533**				-0.061 ns				0.366**	
WUE (W4)				0.429**				-0.191 ns				-0.026 ns

**, *,: Significant at 1 , 5 % level and ns: Not significant respectively, W1, W2, W3 and W4 in parenthesis are water regimes treatment (W1, W2, W3 and W4 = FC, 25, 40 and 60 reduction % of amount of water regimes in FC, respectively)

significant correlation coefficient between WUE and HI under the most severe drought stress also, supported this conclusion.

It is interesting to note here that the relationship between WUE and TDM is not stress-dependent, whereas the relationship between WUE and pod yield and relationship between WUE and HI were stress-dependent. Therefore, care must be taken when compare the results of different experiments. The results are also important for the use of selection conditions for WUE and related traits.

The correlations between WUE and its surrogate traits were presented in Table 7. The correlation between WUE and SCMR showed increasing pattern with the increase in severe drought stress, starting with non-significant correlation under fully-irrigated conditions and becoming significant under all levels of drought stress conditions. The correlation between WUE and canopy temperature showed decreasing pattern in negative direction, starting with negative and significant correlation under well-watered conditions and becoming non-significant correlations under severe drought stress conditions. For the relationship between WUE and SLA, the positive and significant correlation was observed under severe conditions only (W3), where, as there were non-significant correlations under other water conditions, showing inconsistent pattern.

Earlier study has indicated that chlorophyll density has been related to WUE based on measurement of SCMR (Sheshshayee *et al*., 2006). The SCMR is an indicator of the photo-synthetically active light-transmittance characteristics of the leaf, which is dependent on the unit amount of chlorophyll/unit leaf area (chlorophyll density) (Richardson *et al*., 2002). In general, the thicker leaves usually have a higher density of chlorophyll/unit leaf area and hence have a greater photosynthetic capacity compared with thinner leaves. Peanut genotypes with high chlorophyll density have more photosynthetic machinery. Present study demonstrated that drought stress increased WUE as well as increasing SCMR in peanut. Importantly, the variation in WUE was closely correlated with genotype variation in SCMR and hence with photosynthetic capacity. However, there was no relationship in non drought stress, so, it could be used as a potential indicator of WUE under drought conditions. For canopy temperature, it could be used as a potential indicator of WUE under well-water condition, but could not be used under stress drought. For the relationship between WUE and SLA showed not significant correlation and inconsistent pattern. More previous studies found the relationship between SLA and WUE. Wright *et al.* (1994) and Sheshshayee *et al.* (2006) reported the strong negative relationship between WUE and SLA and suggested that the genotypes with lower SLA had higher WUE. The difference in the results might be caused by late sampling date in our study. At harvest, the size of leaves readily reduced, but we could not take samples earlier because of destructive sampling. The appropriate sampling times could be at 60 or 90 days after emergence was fitness than harvest. Nigam and Aruna (2007) suggested that SLA observations under moisture deficit conditions can be recorded at any time after 60 days of the crop growth and Nageswara Rao *et al.* (2001) measured SLA values at 50-60 day-old plants.

14 PI 430238 was the most promising line, showing consistently high TDM, SCMR and low canopy temperature. It also had high WUE under all of stressed conditions, but it had high pod yield only under well-irrigated conditions and mild drought conditions. However, 14 PI 430238 had relatively low HI under all conditions.

Tifton-8 had consistently high TDM and WUE under the four stress treatments and its canopy temperature was low under non-stress conditions only. Unfortunately, Tifton-8 had low pod yield because of low harvest index (Songsri *et al*., 2008b). 205 PI 442925 had consistently high SCMR under four conditions, whereas TDM and WUE consistently high under the first three water regimes. But, pod yield, HI and canopy temperature not showed performance well. KK 60-3 and 101 PI 268659 had performance well TDM, pod yield and WUE under severe drought only, but they did not perform well for other characters. For KKU 60 had consistently high pod yield and SCMR and low canopy temperature under the four water regimes treatment. These peanut genotypes are promising for use as parents in peanut breeding programs for drought resistance.

So, drought stress reduced TDM, pod dry weight, HI, WUE and SLA, but increased SCMR and canopy temperature. WUE had higher contribution to TDM than to pod yield which was dependent on HI. The correlation of WUE was positively related to SCMR for most water regimes except for normal conditions and negatively related to canopy temperature in non-stress and mild stress conditions. SCMR is the most appropriate surrogate trait for WUE. The genotypes with high WUE in all of drought levels were Tifton-8, 14 PI 430238 and 205 PI 442925. KK 60-3, 101 PI 268659 had high WUE under severe drought conditions only. 14 PI 430238 had consistently high TDM, SCMR and low canopy temperature and it had high pod yield only under well-irrigated conditions and mild drought conditions. Tifton-8 had consistently high TDM and WUE under the four stress treatments and its canopy temperature was low under non-stress conditions only. 205 PI 442925 had consistently high SCMR under four conditions, whereas its TDM and WUE consistently were high under the first three water regimes. KK 60-3 and 101 PI 268659 performed well for TDM, pod yield and WUE under severe drought only. The genotypes identified might be useful in future breeding programs for drought tolerance.

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