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Identification of Attributes Contributing to High Temperature Tolerance in Blackgram (*Vigna mungo* L. Hepper) Genotypes

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Abstract: ***Drought and high temperature stresses are the most important constraints to blackgram (Vigna mungo L. Hepper) production and account for about 50 per cent of the yield loss. A field experiment was laid out with seventeen genotypes of blackgram in order to identify genotype/s tolerant to high temperature of above 40°C and quantified the impacts on morphological, seed yield and yield contributing characters. High temperature influenced pod setting, seed filling and significantly reduced the seed yield however there was significant variation in the magnitude of response among the genotypes. Among the seventeen blackgram genotypes, IC 436610 and PU-19 performed better for seed yield and yield contributing traits under both cool and warm seasons with moderate reduction at high temperature. From the present study it is evident that adequate genetic variability exists within the blackgram genotypes which can able to tolerate high temperature of >40°C. The yield contributing characters such as pod number, seed number and 100-seed weight are three key traits, which are useful in screening the genotypes for high temperature tolerance along with better seed yield in blackgram by plant breeders.***

Keywords: High temperature tolerance, genotypic variability, Phenology, Seed number, Seed yield, 100 seed weight

1. Introduction

High temperature stress is the most common adverse environment and has been limiting yield of many crops including blackgram. According to IPCC (2007) predictions by the year 2050, temperature would rise by 3-4°C over current levels. The impacts of climate change will be more pronounced on rainfed crops which account for nearly 60% of cropland area. Major pulse crop production is under marginal soils of rainfed area in semi-arid tracts. Temperature rise impacts are more drastic on short term pulses production as extreme temperatures affect flower and pod setting of these crops. In general, even minute rise in temperature above threshold levels results drastic reduction in seed yield (Summerfield *et al*., 1984; Wang *et al.,* 2006).

Blackgarm (*Vigna mungo (L.)* Hepper) known as urad, mash bean is an important short duration legume crop after chickpea, pigeon pea and soybean. Blackgram contains high level of protein (25%) and it share major protein requirement in vegetarian diet. As a legume crop it is very useful for intercropping, crop rotation to improve soil fertility and can be used as green manure. In India, this crop grown on 3.26 million hectares in 2010-2011, which produced 1.75 million tons grains with an average productivity of Kg ha^{-1} (Anonymous, 2012). To meet the demand of increased population, improvement in pulse production is required.

Black-gram is thermo-sensitive crop and its yield is much sensitive to high temperature of above 35°C which leads to massive flower and pod drop resulting no or low yield. To limit the effects of temperature stress on blackgram, genetic management can be considered as appropriate solution. In this approach temperature stress tolerant genotypes are identified by proceeding thorough screening processes at high temperature stress condition of above 40°C at critical pheno-phages. An understanding of tolerance mechanisms at genotypic level towards high temperature stress facilitates the identification of temperature stress tolerant genotypes with reasonable seed yield. In view of this, the present experiment was conducted to assess the morphological and yield related parameters of seventeen blackgram genotypes under high temperature stress conditions.

2. Materials and Methods

A field experiment was laid out with seventeen blackgram genotypes during 2013-2014 at Hayathnagar Research farm of Central Research Institute for Dryland Agriculture, Hyderabad, situated at 17°18-N latitude, 78°36-E longitude and 515 m above msl. The present study was carried out with seventeen blackgram (*Vigna mungo* L. Hepper) genotypes namely IC 281987, IC 282009, IC 343947, IC 343952, IC 398971, IC 436519, IC 436610, IC 436652, IC 436720, IC 436753, IC 519805, IC 587751, IC 587752, IC 587753, were obtained from NBPGR, Regional station, Hyderabad and with three released popular varieties PU-19, LBG-20 and T-9. In order to identify tolerant genotype/s to high temperature stress, the experiment was sown during rabi (cool) and summer (warm) seasons of 2013-2014 in RBD in three replications with a plot of size 7 x 9 m, spacing of 30cm for row to row and 10 cm for plant to plant.

In order to create two different temperature regimes, the cool season crop was sown in October, 2013 and warm season crop in March, 2014 and maintained stress free by irrigating at regular intervals. All the recommended cultural practices were followed for raising a good field crop. During the cool season, the average temperature of crop growth period was 21.9°C with a minimum of 8.4°C and maximum of 33.2°C, while with warm season, the average temperature was 29.9°C with a minimum of 15.7 °C and maximum of 41.7°C. The crop was exposed to high temperature of above 40°C at flowering and pod setting stage in warm season while it was 30.3°C in cool season (Table 1).

The plants were harvested at maturity and representative plant samples of three replicates of each genotype from each replication were randomly chosen to determine morphological and yield characters. Observations were recorded on plant height (cm), number of branches per plant, days to 50% flowering, number of clusters, pod number, seed number, pod weight (g/pl), seed yield (g/pl) and 100-seed weight (g). The two factor analysis of variance (ANOVA) was carried out for genotypes, temperature levels and their interaction at $P<0.05$ and $P<0.01$ significance level.

3. Results and Discussion

The ANOVA results revealed that plant height, number of clusters, pod number, pod weight, seed number, 100-seed weight and seed yield were highly significant ($p<0.01$) for genotypes, temperature levels and their interaction, while days to 50% flowering and number of branches per plant were highly significant ($p<0.01$) for genotypes, temperature levels and non-significant for genotype x temperature levels (Table 2).

The high temperatures during warm season significantly reduced plant height (30%), number of branches per plant (24%), number of clusters per plant (29%), number pods per plant (42%) and number of seeds per plant (62%), pod weight (62%), 100-seed weight (12.42%) and seed yield (67%) of selected blackgram genotypes as compared with cool season (Table 3a,3b). Though all morphological and yield parameters decreased with high temperature, however there was significant variation in the magnitude of response among the genotypes. Rise in temperature beyond the normal range impairs natural functions of plant metabolism, consequently reduce crop yield. Temperature plays crucial role in plant growth such as germination, seedling growth and reproductive performance (flowering, pod formation and seed filling) (Hasanuzzaman *et al.,* 2013; Chaves *et al.,* 2009). Similar genotypic variability in response to temperature stress also reported in chickpea (Krishnamurthy *et al*., 2011).

During cool season, the plant height ranged from 19.34 cm (IC 343947) to 37.00cm (IC 398971) while with warm season it ranged from 18.12 cm (T-9) to 20.16 cm (IC 519805) and the mean performance was 28.35 to 19.20 cm respectively. There was significant reduction in the plant height of all the selected blackgram genotypes under the warm season over cool season except genotype IC 343947 and the percent of reduction ranged from 1.76% (IC 343947) to 47.23% (IC 398971) with high temperature. The lowest reduction in plant height was observed in genotype IC 343947, where it maintained moderate plant height in both cool and warm season. Kumar *et al.* (2012) reported that temperature tolerant chickpea genotypes recorded lowest reduction in plant height.

Under cool season, the number of branches per plant ranged from 3.33 (IC 281987 and T-9) to 5.67 (IC 436652) while with warm season it ranged from 2.00 (IC 281987) to 4.00 (IC 436753, LBG-20) and the mean performance was 4.28 to 3.23 respectively. The percent of reduction of number of branches per plant ranged from 4.6% (PU-19) to 40% (IC 281987) at high temperature. The genotypes IC 436720 and LBG-20 recorded maximum number of branches/plant under both cool and warm seasons and minimum by IC 281987 and T-9.

Plants escapes from high temperature stress through early flowering and early maturity and these are the two major components of high temperature tolerance. Under cool season, days to 50% flowering was ranged from 32.67 (IC 436720) to 41.67 days (IC 398971) while with warm season it ranged from 30.17 (T-9) to 41.33 days (IC 398971) and the mean performance was 35.85 to 33.52 days respectively. There was significant difference in flowering time among the blackgram genotypes in both cool season and warm season. High temperature influenced the phenology of 50% flowering in selected blackgram genotypes, it was early by five days with IC 587751 and three days with six genotypes- IC 436519, IC 436753, IC 587752, PU-19, LBG-20 and T-9. In the present study, the genotypes- IC 398971 and IC 436720 recorded similar days to 50% flowering in both cool and warm seasons revealing their thermo insensitivity of flowering across temperature regimes. among these the genotype IC 398971 was with highest number of days (42 days) and the genotype IC 436720 was with lowest number of days (33 days) to 50% flowering. Krishnamurthy *et al.* (2011) reported that in chickpea days to 50% flowering was delayed and days to maturity was accelerate at high temperature, however Summerfield *et al.* (1984) and Upadhyaya *et al.* (2011) observed early flowering and forced maturity in chickpea.

The number of clusters per plant reduced significantly under high temperature. Under cool season, the number of clusters per plant was ranged from 13.19 (T- 9) to 25.00 (PU-19) while with warm it ranged from 7.33 (IC 343947) to 17.33 (IC 398971). The percent of reduction of number of clusters per plant ranged from 0.57% (IC 436610) to 54.27% (IC 343947) with high temperature and the mean performance was 17.28 to 12.15 respectively. The genotypes IC 398971, IC 436519, IC 436610 and PU-19 recorded less than 10% of reduction, among them IC 436610 recorded lowest reduction (0.57%) which incidentally maintained maximum number of clusters in both cool and warm season. With high temperature, more than 50% reduction in clusters per plant was recorded with IC 281987 (50.94%), IC 34394 (54.27%), IC 436652 (45.85%), IC 587751 (44.73%), IC 587752 (51.43%) and LBG-20 (34.55%) revealing their sensitivity of yield components to high temperature.

Temperature stress at flowering and pod setting affects seed yield as pod number per plant is main yield contributing trait. Under cool season, the number of pods per plant was ranged from 38.62 (IC 587753) to 45.06 (LBG-20) while with warm season it ranged from 11.00 (IC 587751) to 40.00 (LBG-20). The percent of reduction in number of pods per plant ranged from 3.26% (PU-19) to 74.59% (IC 587751) and the mean performance was 41.60 to 24.18

respectively. The genotype PU-19 with maximum number of pods per plant in both cool and warm seasons also recorded lowest reduction (3.26%) with high temperature. Krishnamurthy *et al*. (2011) and Wang *et al.* (2006) also reported that higher temperature at flowering decreased the yield and yield contributing characters such as pod number, seed number in chickpea genotypes. Among the seventeen blackgram genotypes, IC 436610 and IC 587753 recorded < 20% reduction and these two genotypes performed better for this trait under cool season while moderate at high temperatures. There was more than 80% reduction in pod number with two genotypes IC 587751 and IC 587752 showing their high susceptibility to high temperatures. Devasirvatham *et al.* (2012, 2013) identified heat-tolerant chickpea genotypes by exposing them to more than critical temperature of 37°C which affects the pod set and reduce the crop yield.

High temperature during flowering drastically reduced (>50%) the number of seeds per plant of majority blackgram genotypes and it could be due to sensitivity of their pollen to high temperature which in turn affects the pollination and seed set (Devasirvatham *et al.,* 2013). Under cool season, the number of seeds per plant ranged from 180.02 (IC 519805) to 255.37 (LBG-20) while with warm season (summer) it ranged from 42.00 (IC 587752) to 161.33 (PU-19) and the mean performance was 210.33 to 80.73 respectively. The percent of reduction of number of seeds per plant ranged from 25.46% (PU-19) to 81.04% (IC 587752) with high temperature. Among the seventeen blackgram genotypes, PU-19 was able to maintain high yield potential under both seasons with lowest reduction (25.46%). The genotypes IC 398971, IC 436610, IC 587753 and PU-19 registered <50% reduction in seed number with high temperature. In chickpea genotypes also the reduction of plant biomass and number of seeds per plant under high temperature was reported (Upadhyaya *et al.,* 2011; Wang *et al*., 2006; Kiran and Chimmad 2015).

The response of pod weight at high temperature followed similar trend as number of pods per plant. Under cool season, the pod weight ranged from 14.94 g/pl (T-9) to 17.95 g/pl (LBG-20) while with warm season it ranged from 3.10 g/pl (IC 587751) to 11.43 g/pl (PU-19) and the mean performance was 16.42 g/pl to 6.22 g/pl respectively. The percent of reduction of pod weight ranged from 31.08% (PU-19) to 80.74% (IC 587751) with high temperature. Under cool season the genotype LBG-20 recorded maximum pod weight followed by IC 281987, IC 343952, IC 398971 and IC 436610 while with high temperature at flowering, PU-19 recorded maximum pod weight followed by IC 436610 and minimum with IC 587751. The percent of reduction was <50% in all genotypes except PU-19 (31.08%), IC 4336610 (41.10%) and IC 383971 (45.5%) and these genotypes performed well in both cool and warm seasons. Similar responses in groundnut were reported with high temperature (Prasad *et al.,* 2001).

Seed yield reduced under high temperature compared to their cool season. Under cool season, the seed yield ranged from 8.35 g/pl (T-9) to 11.82 g/pl (IC 436720) while with warm season it ranged from 1.71 g/pl (T-9) to 6.73 g/pl (PU-19). The percent of reduction of seed yield ranged from 37.91% (PU-19) to 82.02% (IC 587751) with high temperature. The mean performance of seed yield under cool and warm seasons was 10.53 g/pl to 3.57 g/pl respectively. High temperature at flowering stage reduced the number of pods per plant, pod weight (g/pl), number of seeds per plant and seed weight (g/pl) as compared to cool season. Though genotypes IC 398971 (11.80 g/pl), IC 436610 (11.69 g/pl) maintained similar seed yield under cool season, the reduction in seed yield of these genotypes varied under high temperature and it was 52.86%, 46.50% and 72.78% respectively, revealing the variability in response to high temperature among these high yielding genotypes. The reduction in seed yield with high temperature at flowering was < 50% in majority of the genotypes, except PU-19 (37.91%) and IC 436610 (46.50%) and incidentally these two genotypes also recorded highest seed yield under both seasons. Wang *et al.* (2006) recorded decreased seed yield through reduced number of seeds per plant and seed weight during high temperature in chickpea. Our present results are in accordance with the findings of Basu *et al*. (2009), Devasirvatham *et al*. (2010) and Gaur *et al*. (2015) in chickpea.

Impact of high temperatures also registered for seed filling by reducing the 100-seed weight. Under cool season, the 100-seed weight ranged from 4.10 g (T-9) to 5.81 g (IC 343947) while with warm season it ranged from 3.26 g (T-9) to 4.73 g (IC 436610) and the mean performance was 4.95 g to 4.33 g respectively. The percent of reduction of 100-seed weight ranged from 3.08% (IC 519805) to 21.39% (IC 436720) with high temperature. 100-seed weight reduced in all the genotypes as high temperature impairs the translocation of photosynthates and reduced the seed size (Prasad *et al.,* 2008). The three genotypes- IC 343952, IC 519805 and IC 587752 recorded less than 5% of reduction in 100-seed weight with high temperature while genotype T-9 (20.4%) recorded high percent of reduction. Adam *et al.* (2013) also reported that high temperature at reproductive stage decreased the seed yield and 100-seed weight in soya bean genotypes.

4. Conclusion

High temperature significantly reduced the blackgram plant height, days to 50% flowering, number of branches, number of clusters, number of pods, number of seeds, pod weight, seed yield and 100-seed weight of all genotypes, however the magnitude of response varied significantly with individual genotype as well as with each trait. The two blackgram genotypes IC 436610 and PU-19 performed better for seed yield and yield contributing traits under both cool and warm seasons with moderate reduction at high temperature. The genotype PU-19 maintained number of pods, less reduction in seed number while IC 436610 recorded less reduction in 100 seed weight, revealing the different strategies adopted by these genotypes to maintain the better yield at high temperature stress. The present results clearly indicating that sufficient genetic variability exists within the blackgram genotypes in terms of tolerance to high temperature of >40°C. The yield contributing characters such as pod number, seed number and 100-seed weight are three key traits, which are useful in screening the genotypes having tolerance to high temperatures along with better seed yield and can be adopted by plant breeders to improve the heat tolerance in blackgram.

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Table 1: Temperatures at different phenophases of blackgram during cool and warm season

Temperature (ºC)				
Seasons	Growth Stages	Min.	Max.	Average
Cool season	Vegetative Stage	14.4	33.2	24.4
	Flowering Stage	10.2	30.4	21.2
	Pod setting Stage	8.4	29.7	19.3
	Total crop growth period	**8.4**	**33.2**	**21.9**
Warm season	Vegetative Stage	19.5	39.4	30.3
	Flowering Stage	15.7	41.7	30.3
	Pod setting Stage	18.0	39.6	28.8
	Total crop growth period	**15.7**	**41.7**	**29.9**

Table 2: ANOVA for yield contributing parameters in cool and warm season

Parameters	Genotypes (G) df (16)	Temperature level (T) df (1)	Genotypes × Temperature levels (G × T) df (16)	Error df (66)	CV%
Plant height (cm)	42.61**	2136.66**	42.94**	8.522**	12.28**
Day to 50 % Flowering	31.34**	165.68**	NS	2.31**	4.33**
No. of branches/ plant	2.56***	27.95**	NS	0.448**	17.82**
No. of clusters/ plant	31.63**	670.87**	19.52**	7.30**	18.36**
Number of pods/ pl	94.43**	7737.96**	115.38**	42.49**	19.82**
Number of seeds/ pl	3004.45**	428328.0**	1904.10**	613.29**	17.02**
Pod weight (g/pl)	211.49**	2652.77**	8.65**	2.19**	13.07**
100-seed weight (g)	728.2**	607.36**	739.81**	1.30**	13.88**
Seed yield (g/pl)	7.13**	1236.3**	2.496**	0.905**	13.49**

** Significant at (p<0.01) level; NS- Non-significant

Table 3(a): Yield and yield contributing characters in seventeen blackgram genotypes in cool and warm season and % of reduction

Genotypes	Plant height (cm)			No. of branches			Days to 50 % Flowering			No. of clusters/pl			No. of pods/pl		
	CS	WS	% Red	CS	WS	% Red	CS	WS	Difference	CS	WS	% Red	CS	WS	% Red
IC 281987	31.67	18.83	40.53	3.33	2.00	40.00	35.5	33.5	2.00	18.34	9.00	50.94	43.33	15.33	64.62
IC 282009	29.33	18.68	36.32	5.00	3.78	24.47	35.5	33.67	1.83	17.20	15.00	12.79	42.24	33.00	21.87
IC 343947	19.34	19.00	1.76	3.67	3.33	9.09	34.17	31.5	2.67	16.04	7.33	54.27	41.33	16.33	60.48
IC 343952	25.67	19.20	25.19	4.00	3.00	25.00	34.83	32.5	2.33	18.25	11.67	36.06	43.17	19.67	54.44
IC 398971	37.00	19.53	47.23	3.82	2.33	38.86	41.67	41.33	0.34	18.34	17.33	5.51	43.33	32.33	25.38
IC 436519	30.67	18.83	38.59	3.82	2.67	30.13	36	32.67	3.33	14.44	14.33	0.72	39.17	24.33	37.87
IC 436610	30.33	20.09	33.78	5.00	3.82	23.67	35.5	34.17	1.33	17.10	17.00	0.57	42.53	33.67	20.84
IC 436652	34.33	18.31	46.66	5.67	3.50	38.24	35.33	33.17	2.16	16.62	9.00	45.85	41.63	17.33	58.36
IC 436720	28.00	19.83	29.19	3.55	3.33	6.10	32.67	32.5	0.17	16.67	13.58	18.54	38.72	28.00	27.69
IC 436753	33.33	18.41	44.76	5.33	4.00	25.00	36.33	33.17	3.16	16.65	9.00	45.96	41.71	23.67	43.26
IC 519805	24.33	20.16	17.16	5.00	3.68	26.33	35.5	34.17	1.33	13.58	10.33	23.89	38.72	23.00	40.60
IC 587751	23.67	19.48	17.69	3.75	3.33	11.11	38.33	32.83	5.50	18.09	10.00	44.73	43.30	11.00	74.59
IC 587752	27.67	19.19	30.63	3.50	2.67	23.81	36.5	33.5	3.00	18.53	9.00	51.43	43.30	13.67	68.43
IC 587753	22.33	19.16	14.21	5.33	3.33	37.50	34.67	34	0.67	15.67	13.68	12.68	38.62	31.67	18.00
PU-19	36.67	19.51	46.80	3.67	3.50	4.55	36	33	3.00	25.00	16.36	34.55	41.35	40.00	3.26
LBG-20	27.00	20.05	25.73	5.00	4.00	20.00	37.5	34	3.50	20.06	11.67	41.85	45.06	26.00	42.29
T-9	20.67	18.12	12.31	3.33	2.67	20.00	33.5	30.17	3.33	13.19	12.33	6.47	39.64	22.00	44.50
Mean	28.35	19.20	29.9	4.28	3.23	23.8	35.85	33.52	2.3	17.28	12.15	28.63	41.60	24.18	41.56
Min	19.34	18.12	1.76	3.33	2.00	4.55	32.67	30.17	0.17	13.19	7.33	0.57	38.62	11.00	3.26
Max	37.00	20.16	47.23	5.67	4.00	40.00	41.67	41.33	5.50	25.00	17.33	54.27	45.06	40.00	74.59

CS: cool season; WS: warm season; % Red- % of reduction during warm season over cool season

Table 3 (b): Yield and yield contributing characters in seventeen blackgram genotypes in cool and warm season

Genotypes	**No.of seeds/pl**			**Pod weight (g/pl)**			**Seed yield (g/pl)**			**100 seed weight (g/pl)**		
	CS	WS	% Red	CS	WS	% Red	CS	WS	% Red	CS	WS	% Red
IC 281987	197.56	45.33	77.05	17.25	3.39	80.34	9.93	1.92	80.70	4.76	4.23	11.30
IC 282009	197.22	89.33	54.70	16.87	8.34	50.58	10.66	4.15	61.06	5.06	4.65	8.13
IC 343947	222.67	65.33	70.66	15.88	5.94	62.58	10.87	3.80	65.08	5.81	4.69	19.26
IC 343952	241.67	64.33	73.38	17.27	4.87	71.80	11.25	2.81	75.04	4.61	4.37	5.20
IC 398971	245.00	123.33	49.66	17.23	9.39	45.51	11.80	5.56	52.86	4.81	4.51	6.32
IC 436519	192.00	73.67	61.63	16.15	6.37	60.55	11.08	3.48	68.59	5.49	4.72	13.96
IC 436610	226.67	132.33	41.62	17.15	10.10	41.10	11.69	6.26	46.50	5.24	4.73	9.79
IC 436652	192.75	67.00	65.24	16.69	4.87	70.83	9.61	2.77	71.14	4.89	4.14	15.25
IC 436720	208.80	73.67	64.72	15.69	5.78	63.13	11.82	3.22	72.78	5.56	4.37	21.39
IC 436753	182.73	81.00	55.67	16.48	6.40	61.18	9.75	3.80	61.01	5.38	4.69	12.82
IC 519805	180.02	66.33	63.15	15.23	5.40	64.56	10.55	3.12	70.41	4.86	4.71	3.08
IC 587751	218.90	45.67	79.14	16.08	3.10	80.74	10.40	1.87	82.02	4.90	4.09	16.49
IC 587752	221.50	42.00	81.04	16.16	3.17	80.36	9.38	1.82	80.59	4.34	4.20	3.22
IC 587753	194.57	108.00	44.49	15.54	7.79	49.88	9.81	4.64	52.68	4.77	4.30	9.98
PU-19	216.43	161.33	25.46	16.58	11.43	31.08	10.84	6.73	37.91	4.90	4.17	14.92
LBG-20	255.37	81.33	68.15	17.95	5.50	69.37	11.27	3.04	73.00	4.65	3.74	19.59
T-9	181.73	52.33	71.20	14.94	3.90	73.90	8.35	1.71	79.55	4.10	3.26	20.40
Mean	**210.33**	**80.73**	**61.59**	**16.42**	**6.22**	**62.21**	**10.53**	**3.57**	**66.53**	**4.95**	**4.33**	**12.42**
Min	**180.02**	**42.00**	**25.46**	**14.94**	**3.10**	**31.08**	**8.35**	**1.71**	**37.91**	**4.10**	**3.26**	**3.08**
Max	**255.37**	**161.33**	**81.04**	**17.95**	**11.43**	**80.74**	**11.82**	**6.73**	**82.02**	**5.81**	**4.73**	**21.39**

CS: cool season; WS: warm season; % Red- % of reduction during warm season over cool season