

## Combining Ability Analysis of beta-Carotene, Total Carotenoids and Other Grain Characteristics in Pearl Millet\*

B.S. Khangura, K.S. Gill and P.S. Phul

Department of Plant Breeding, Punjab Agricultural University, Ludhiana (India)

**Summary.** Combining ability studies with respect to grain quality characteristics viz., beta-Carotene, total carotenoids, protein content, 250-grain weight, grain hardness and grain yield were carried out from a 13 × 13 diallel cross set in pearl millet [*Pennisetum typhoides* (Burm S&H)]. The parents versus hybrids comparison indicated significant heterosis for all the traits under study. In general the hybrids having higher grain yield had bold hard grains with more carotene but low protein content, although a few hybrids combined high yield with an average protein percentage. The relative proportions of the general and specific combining ability variances indicated predominance of non-additive genetic variance with respect to all the traits. The per se performance of parents provided a fairly good indication of their combining ability in most cases. Parents possessing desirable grain quality characteristics were identified. Breeding implications are discussed.

**Key words:** Combining ability – beta-Carotene – Carotenoids – Protein Content – Grain characteristics – Pearl millet

### Introduction

Pearl millet (*Pennisetum typhoides* (Burm S&H)) is an important food crop occupying about 12 million hectares in India. It is extensively grown in dry areas and is a diet staple of a vast population in the country. Its grain is very rich nutritionally and contains a higher protein content than many other cereals. Besides protein, pearl millet

grains also contain sufficient amount of beta-Carotene which is the precursor of vitamin A. Thus, it can serve as an additional source of vitamin A (Curtis et al. 1966).

Although grain yield of this crop has been considerably increased by the development of high yielding hybrids (Athwal 1965; Gill et al. 1975), attempts to improve its nutritive value have been rather limited. In addition to chemical composition, the physical attributes of a grain, such as colour, size and hardness, are also important constituents of grain quality and may enhance its market value. Therefore, concerted efforts are required to bring about simultaneous improvement in grain yield and quality.

The present study was undertaken to evaluate some inbred lines of pearl millet with yellow endosperm for combining ability with respect to quality characteristics and grain yield from a 13 × 13 diallel cross set.

### Materials and Methods

Thirteen inbred lines, viz., PIB-197, PIB 11-1, PIB 195, PIB 274, PIB 51-1, PIB 31, PIB 128, Pb 103B, Pb 111B, PIB 146-1, PIB 703-1, Tift 239B and DYG 39 were crossed in all possible one-way combinations to yield 78 crosses. Except for five of the lines, viz. Pb 103B, Pb 111B, PIB 146-1, PIB 703-1 and Tift 239B, all the others had varying shades of yellow endosperm colour. Ninety-one progenies, comprising 13 parents and 78 F<sub>1</sub>s, were grown in a randomized complete block design with 3 replications, each at two locations in the Punjab (Ludhiana and Faridkot) from July to October, 1973.

Total carotenoids were extracted with 40 ml of 1:1 N-hexane: acetone from a 10 gm sample. The samples were kept overnight, filtered and then washed with extractants until all the pigments were collected in the filtrate. The volume was made up to 50 ml with the solvent. A part of this solution was used to measure the absorbance in Spectronic-20 at 430 m $\mu$ . The total carotenoids were estimated in ug/100 gm from the standard curve (Fig. 1).

For estimating the beta-Carotene by column chromatography, 25 ml of the filtrate was used. Acetone was removed by washing the filtrate with water 2-3 times. The measured amount of stan-

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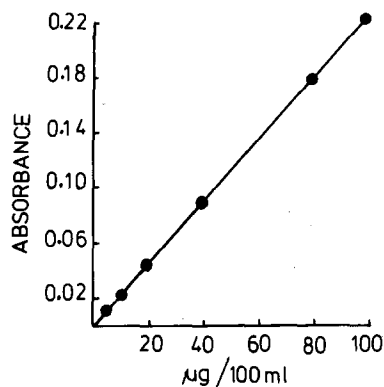


Fig. 1. Standard curve for estimating total carotenoids

Standard carotene solution was then added as an internal standard. The sample as well as the standard solution were passed through a column made up of equal parts of neutral Alumina Buckman Activity-1 (activated) and sodium sulphate. The length of the column was approximately 10 cm and the breadth 1.2 cm. The elution was done with 3% acetone in N-hexane until the beta-Carotene was eluted leaving behind the other pigments in the column. The volume was made up to 50 ml with N-hexane. The absorbance of the sample as well as of the standard was measured in the Spectronic-20 at 430 m $\mu$ . The difference in absorbance was noted and the amount of beta-Carotene in the sample was determined in  $\mu$ g/100 gm from the standard curve.

Protein content was determined by the method suggested by Mackenzie and Wallace (1954). 250-grain weight was taken as an index of grain size. Grain hardness was measured on the hardness tester as weight in kg required for breaking the grain on 10 random grains selected from the produce of each plant and average weight was recorded. Grain yield per plant was recorded in grams.

The combining ability analysis was carried out according to model 1 method 2 of Griffing (1956). Correlation coefficients were worked out among the quality traits themselves, as was also done with grain yield.

## Results and Discussion

### Average Performance of Parents and Hybrids

The analysis of variance showed highly significant differences among parents as well with the hybrids for various characteristics determining grain quality and yield (Table 1). The parents differed significantly from the hybrids with respect to all the characters, indicating the presence of significant heterosis for these characters. An examination of the overall mean of the parents and the hybrids (Table 2) revealed that the heterosis for beta-Carotene, total carotenoids and protein content was in the negative direction. Mahadevappa (1967), Phul et al. (1969), Harinarayana and Murty (1970) and Phul et al. (1973) also reported negative heterosis for protein content. Worzella et al. (1965) observed similar results for beta-Carotene in sorghum. However, for grain hardness, 250-grain weight and grain yield, the heterosis was in the desirable direction with the hybrids having, in general, higher grain yield and bold, hard and yellow coloured grains. The carotene content of the grains was also higher but the protein content was low in most cases. For example, the highest yielding hybrid, PIB 128  $\times$  Pb 111B, had

Table 1. Analysis of variance for design of experiment for five grain quality traits and grain yield in pearl millet

Source of variation	d.f.	250-grain weight	Grain hardness	Protein content	Total carotenoids	betacarotenoids	Grain yield
<i>Ludhiana</i>							
Replications	2	0.00	10.74 <sup>a</sup>	88.94 <sup>a</sup>	83618.60 <sup>a</sup>	788.41 <sup>a</sup>	1008.05 <sup>b</sup>
Progenies	90	0.36 <sup>a</sup>	2.52 <sup>a</sup>	8.08 <sup>a</sup>	5107.19 <sup>a</sup>	130.86 <sup>a</sup>	1322.05 <sup>a</sup>
Parents	12	0.25 <sup>a</sup>	5.14 <sup>a</sup>	6.89 <sup>a</sup>	8232.43 <sup>a</sup>	221.66 <sup>a</sup>	386.46
Hybrid	77	0.25 <sup>a</sup>	1.81 <sup>a</sup>	7.48 <sup>a</sup>	3015.68 <sup>a</sup>	97.38 <sup>b</sup>	884.66 <sup>a</sup>
Parents versus hybrids	1	10.63 <sup>a</sup>	25.18 <sup>a</sup>	68.67 <sup>a</sup>	128650.77 <sup>a</sup>	1619.68 <sup>a</sup>	46228.13 <sup>a</sup>
Error	180	0.04	0.59	1.58	1156.98	57.75	311.97
<i>Faridkot</i>							
Replications	2	0.09	6.05 <sup>a</sup>	6.92 <sup>b</sup>	16000.80 <sup>a</sup>	1198.81 <sup>a</sup>	1018.11 <sup>a</sup>
Progenies	90	0.32 <sup>a</sup>	2.03 <sup>a</sup>	8.60 <sup>a</sup>	5102.98 <sup>a</sup>	141.60 <sup>a</sup>	413.31 <sup>a</sup>
Parents	12	0.28 <sup>a</sup>	2.50 <sup>a</sup>	6.66 <sup>a</sup>	9258.37 <sup>a</sup>	327.57 <sup>a</sup>	301.62 <sup>b</sup>
Hybrids	77	0.22 <sup>a</sup>	1.49 <sup>a</sup>	8.81 <sup>a</sup>	3201.76 <sup>a</sup>	104.40 <sup>b</sup>	306.03 <sup>a</sup>
Parents versus Hybrids	1	8.47 <sup>a</sup>	38.21 <sup>a</sup>	15.59 <sup>a</sup>	101631.95 <sup>a</sup>	774.42 <sup>a</sup>	10013.73 <sup>a</sup>
Error	180	0.04	0.52	2.08	1411.70	64.71	152.06

<sup>a</sup> Significant at 1%  
<sup>b</sup> Significant at 5%

**Table 2.** Mean values and range of variability for some grain quality characteristics and grain yield in pearl millet at Ludhiana and Faridkot

Character	Mean		Range	
	Parents	Hybrid	Parents	Hybrid
<i>Ludhiana</i>				
beta-Carotene ( $\mu\text{g}/100\text{ gm}$ )	38.1	29.6	22.6- 62.6	17.9- 50.3
Total carotenoids ( $\mu\text{g}/100\text{ gm}$ )	228.5	152.5	117.8-339.8	84.8-276.8
Protein content (%)	14.0	12.6	12.1- 17.3	9.1- 16.2
Grain hardness (kg)	4.6	5.4	3.4- 8.3	3.4- 7.0
250-grain weight (gm)	1.4	2.0	0.8- 2.0	1.1- 2.5
Grain yield (gm)	23.7	60.9	10.8- 46.7	21.3-107.3
<i>Faridkot</i>				
beta-Carotene	35.7	29.9	19.1- 62.5	13.5- 48.8
Total carotenoids	226.9	159.4	147.8-443.2	93.1-293.9
Protein content	13.9	13.2	11.5- 17.3	9.8- 18.9
Grain hardness	5.0	6.1	3.9- 7.6	4.2- 7.7
250-grain weight	1.7	2.2	0.9- 2.1	1.5- 2.7
Grain yield	19.9	37.2	6.8- 41.7	14.8- 71.8

higher beta-Carotene (36.7  $\mu\text{g}/100\text{ gm}$ ) and bold, hard grains (grain weight = 2.25 gm and hardness = 6.4 kg) of yellow colour. However, its protein content was below average (9.22%). A similar situation was observed with respect to the second highest yielding hybrid, PIB 128  $\times$  PIB 703-1. However, several hybrids combined high grain yield with average protein content. For example, PIB

146-1  $\times$  PIB 703-1 had a grain yield of 83 gm per plant with more than 14% protein. Similarly, PIB 197  $\times$  PIB 51-1 had a high grain yield (73 gm/plant) with a high protein content (14.25%). Likewise, the hybrid PIB 51-1  $\times$  Tift 239B had high protein content (15.05%) with high carotene (35.7  $\mu\text{g}/100\text{ gm}$ ). Also, the hybrid, Pb 111B  $\times$  PIB 145-1, combined high protein (14.47%) with high beta-Carotene (48.3  $\mu\text{g}/100\text{ gm}$ ). Therefore, desirable combinations were available in the material under study which offers a scope for the simultaneous improvement of these traits.

### Combining Ability Analysis

The choice of the parents is often a major problem for the breeder in the development of superior hybrid varieties. The per se performance is not always a good indication of their superior combining ability. It is the common experience of the breeders that certain combinations nick well to produce superior hybrids whereas the others involving equally promising parents produce disappointing progeny. The combining ability thus depends upon the complex interaction among the genes of the parents. It becomes important then to evaluate the parents for their combining ability.

The analysis of variance for combining ability (Table 3) revealed highly significant differences among general as well as specific combining ability effects at both locations. The estimates of the specific and general com-

**Table 3.** Analysis of variance for combining ability

Source of variation	d.f.	250-grain weight	Grain hardness	Protein content	Total carotenoids	beta-Carotene	Grain yield
<i>Ludhiana</i>							
General combining ability	12	0.23 <sup>a</sup>	2.68 <sup>a</sup>	11.03 <sup>a</sup>	8546.08 <sup>a</sup>	153.28 <sup>a</sup>	653.98 <sup>a</sup>
Specific combining ability	78	0.11 <sup>a</sup>	0.55 <sup>a</sup>	1.41 <sup>a</sup>	1631.66 <sup>a</sup>	51.92 <sup>a</sup>	407.88 <sup>a</sup>
Error	180	0.01	0.19	0.53	578.49	28.88	103.99
<i>Faridkot</i>							
General combining ability	12	0.20 <sup>a</sup>	1.17 <sup>a</sup>	11.48 <sup>a</sup>	7597.38 <sup>a</sup>	139.44 <sup>a</sup>	174.95 <sup>a</sup>
Specific combining ability	78	0.09 <sup>a</sup>	0.60 <sup>a</sup>	1.54 <sup>a</sup>	1775.19 <sup>a</sup>	60.24 <sup>a</sup>	131.90 <sup>a</sup>
Error	180	0.01	0.17	0.69	705.35	32.35	50.69

<sup>a</sup> Significant at 1%

$\sigma^2_{\text{sca}}$	L	0.10	0.36	0.88	1053.17	23.04	303.89
	F	0.08	0.43	0.85	1069.34	27.89	81.21
$\sigma^2$	L	0.007	0.14	0.64	460.96	6.76	16.41
	F	0.008	0.04	0.66	388.14	5.28	2.94

binning ability variance ( $\sigma_{sca}^2$  and  $\sigma_{gca}^2$ , respectively) showed that the former had a higher magnitude than the latter for all six characters under study. This indicates that non-additive gene action was more important with respect to the quality characteristics and grain yield.

The estimates of the general combining ability (g c a) effects (Table 4) of the parents showed that PIB 128 and Pb 111B were better combiners for beta-Carotene, total carotenoids, grain yield and grain hardness. PIB 128 also contributed favourable alleles for grain weight. PIB 197 was a good combiner for protein content. Likewise, PIB 11-1 had a concentration of favourable alleles for grain weight and grain hardness. Since each of these parents possessed one or more of the desirable characters under study, an intermating population involving all possible crosses among them may be synthesized in order to release genetic variability. Selection for desirable combina-

tions of traits in such a population would result in a population of lines combining superior grain quality with high productivity, as also suggested by Ahmed, Murty and Harinarayana (1972). In addition to these lines, Tift 239D was a good combiner for protein content.

The per se performance of the parents provided a fairly good indication of their combining ability in most of the cases (Table 4). PIB 128 and Pb 111B had high per se performance for total carotenoids and beta-Carotene and also high general combining ability. Similarly, PIB 51-1 possessed the highest protein content and high general combining ability effect. Likewise, PIB 274 had a high yield as well as high combining ability. However, this was not always true. For example, PIB 128 had the lowest per se performance but it was the best general combiner for grain yield. On the other hand, PIB 197 and PIB 195 possessed higher grain yields and poor combining ability

Table 4. General combining ability effects and mean values (in parentheses) of the 13 inbred lines for five quality traits and grain yield in pearl millet

Inbreds	beta-Carotene		Total carotenoids		Protein content		Grain hardness		250-grain weight		Grain yield	
	L	F	L	F	L	F	L	F	L	F	L	F
1	2	3	4	5	6	7	8	9	10	11	12	13
PIB 197	0.42 (44.25)	-2.64 (43.20)	-7.26 (285.25)	-15.37 (208.25)	0.51 (15.28)	0.47 (14.05)	-0.07 (4.47)	-0.10 (5.47)	-0.07 (1.40)	-0.08 (1.82)	1.25 (46.67)	1.79 (26.67)
PIB 11-1	-4.06 (31.65)	-3.77 (23.65)	-46.28 (157.00)	-31.10 (167.00)	-0.24 (12.60)	-0.85 (12.13)	0.43 (4.77)	0.11 (5.10)	0.73 (2.03)	0.11 (2.04)	-2.97 (16.67)	-0.51 (12.67)
PIB 195	-2.34 (31.65)	-2.72 (27.65)	-12.56 (218.75)	-6.26 (186.85)	0.06 (15.05)	0.20 (14.42)	-0.52 (4.20)	-0.31 (4.33)	0.05 (1.56)	-0.01 (1.63)	3.39 (38.00)	-0.80 (35.00)
PIB 274	-0.13 (31.95)	0.74 (27.65)	0.13 (218.85)	-3.43 (196.75)	-0.26 (12.13)	0.11 (14.58)	-0.52 (3.90)	-0.37 (4.53)	-0.10 (1.39)	-0.05 (1.83)	6.10 (36.67)	3.48 (41.67)
PIB 51-1	1.14 (36.45)	-0.17 (33.20)	-2.37 (277.00)	-10.26 (186.70)	1.17 (17.27)	1.12 (17.27)	-0.03 (5.20)	-0.04 (5.20)	-0.09 (1.62)	-0.14 (1.39)	0.25 (23.67)	2.43 (9.67)
PIB 31	1.11 (48.20)	1.99 (47.50)	1.24 (230.25)	2.33 (265.50)	0.01 (14.67)	-0.47 (13.30)	-0.20 (3.63)	0.17 (4.83)	0.12 (1.44)	0.22 (2.08)	-7.55 (16.27)	-2.18 (16.33)
PIB 128	5.62 (62.60)	4.95 (62.50)	33.69 (300.35)	24.89 (317.20)	-1.22 (14.67)	-0.78 (14.12)	-0.01 (3.37)	0.37 (5.33)	-0.05 (0.077)	0.10 (1.79)	14.74 (10.00)	5.51 (14.00)
Pb 103B	-4.42 (22.60)	-4.13 (19.10)	-15.17 (117.75)	-13.01 (168.65)	-0.51 (12.37)	-0.49 (12.43)	-0.26 (3.50)	-0.24 (5.13)	-0.11 (1.59)	-0.17 (1.48)	-2.78 (28.00)	-0.29 (23.00)
Pb 111B	6.25 (48.75)	5.91 (53.35)	45.92 (339.75)	48.99 (443.25)	-0.83 (12.60)	-0.27 (14.47)	0.32 (5.13)	0.26 (4.53)	-0.05 (1.22)	0.01 (1.66)	5.44 (27.00)	2.24 (20.00)
PIB 146-1	-0.68 (32.70)	0.08 (32.55)	-3.17 (173.50)	29.94 (238.75)	0.79 (14.12)	1.01 (13.30)	0.21 (5.20)	-0.11 (3.87)	0.15 (1.41)	0.01 (0.95)	-7.85 (15.40)	-4.89 (6.77)
PIN 703-1	0.56 (38.50)	0.78 (38.85)	27.45 (273.50)	6.04 (244.50)	-0.19 (15.05)	-0.26 (15.52)	1.03 (8.33)	0.57 (7.60)	-0.18 (1.48)	-0.14 (1.81)	0.93 (24.53)	2.09 (22.00)
Tift 239B	-2.43 (28.40)	-1.09 (29.55)	-9.39 (204.50)	-11.22 (178.50)	1.71 (14.67)	1.65 (13.65)	-0.11 (4.07)	-0.16 (4.30)	0.14 (1.28)	0.05 (1.38)	-0.33 (15.53)	-6.50 (11.67)
DYG 39	-1.03 (36.20)	0.06 (24.10)	-12.25 (173.50)	-21.53 (147.85)	-1.01 (12.54)	-1.41 (11.55)	-0.28 (3.60)	-0.14 (5.20)	-0.02 (1.20)	0.09 (1.82)	-1.63 (10.00)	3.08 (19.67)
S.E. gi	1.33	1.41	5.96	6.59	0.18	0.21	0.11	0.10	0.03	0.03	2.53	1.76

Table 5. Best performing and best combining parents for various characters

No.	Character	Best performing parents		Best combining parents	
		Ludhiana	Faridkot	Ludhiana	Faridkot
1	beta-Carotene	PIB 128 Pb 111B	PIB 128 Pb 111B	Pb 111B PIB 128	Pb 111B PIB 128
2	Total carotenoids	Pb 111B PIB 128	Pb 111B PIB 128	Pb 111B PIB 128	Pb 111B PIB 146-1
3	Protein content	PIB 51-1 PIB 197	PIB 51-1 PIB 703-1	Tift 239B PIB 51-1	Tift 239B PIB 51-1
4	Grain hardness	PIB 703-1 PIB 146-1	PIB 703-1 PIB 128	PIB 703-1 PIB 11-1	PIB 703-1 PIB 128
5	250-grain weight	PIB 11-1 PIB 51-1	PIB 31 PIB 11-1	PIB 11-1 PIB 146-1	PIB 31 PIB 11-1
6	Grain yield/plant	PIB 197 PIB 195	PIB 274 PIB 195	PIB 128 PIB 274	PIB 128 PIB 274

effects. Similar results have been reported by Govil and Murty (1973). Thus, both g.c.a. as well as the per se performance may be taken into account when selecting the parents for a hybrid breeding programme, as suggested by Ahmad and Murty (1972).

An examination of the per se performance and the specific combining ability (sca) effects of the hybrids indicated that the hybrids having high sca effects may not necessarily have higher per se performance. For example, the combination PIB 146-1 × PIB 703-1 had the highest sca effect for grain yield but it was seventh with respect to its per se performance. On the other hand, the highest yielding hybrid (PIB 128 × Pb 111B) was fourth with respect to its sca effect. Similar results were obtained for other characters. The crosses between two good general combiners were not always good for specific combining ability. For example PIB 174 × PIB 128 involved good combiners for grain yield but its sca effect was not significant. Similarly, Pb 111B × PIB 128 included high combining lines for beta-Carotene, but its sca effect was negative. A similar situation was observed for other characters. Bains et al. (1967) also observed that two high combining lines may not always result in high sca effects. However, the per se performance of the hybrids involving good combiners was generally high, indicating that combining ability of the parents was more related to the per se performance than to the sca effect of their hybrids. This was further confirmed by the fact that the best performing hybrids invariably involved at least one good general combiner (Table 5). This was not true with the hybrids having highest sca effects. The crosses between poor combiners had high sca effects, although their per se performance was not very high. The breeder, however, is interested in the per se performance of the hybrids and not merely

their sca effects. So the per se performance of the hybrids rather than their sca effect is more important for the choice of superior hybrids. Singh et al. (1974) also reported that there was no correlation between the mean performance of the hybrids and their sca effects. It suggests that the high sca effect may be due to genetic diversity among the parents involved in a cross. The mean performance of a cross largely depends on the general combining ability of the parents and may deviate from this expectation because of specific effects resulting from intra- and intergenetic interactions.

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Dr. B.S. Khangura  
Department of Plant Breeding  
Punjab Agricultural University  
Ludhiana (India)