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Protein Quality Traits and their Relationships with Yield and Yield Components of Opaque-2 and Analogous Normal Maize Hybrids and Inbred Lines

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Summary. Diallel cross hybrids obtained by crossing six opaque-2 (o_2) converted inbred lines were compared with a similar series of crosses made between the analogous normal inbred lines together with the parents, for a number of protein quality and oil traits. The results obtained show that there is only a minor decrease in percent protein or yield of protein per hectare caused by the presence of the o_2 gene in recessive homozygous condition, compared with its normal counterpart. On the other hand, percent lysine in the whole kernel, lysine yield per hectare and percent lysine in protein are increased by 53, 45 and 55 percent, respectively, in the o_2 inbred lines and hybrids compared with their normal analogues. Percent oil and oil yield remain almost unaltered by the presence or absence of the o_2 gene. As well as lysine, the content of other amino acids, such as aspartic acid, arginine, glycine, threonine, valine and histidine are also, in general, increased by the presence of the o_2 gene in recessive homozygous condition.

The results obtained have shown that a number of correlation coefficients between the protein quality traits and yield components related to kernel characteristics are negative and significant, especially in the presence of the o_2 gene in recessive homozygous condition. Only two correlation coefficients have been found that could be used with advantage by opaque-2 maize breeders. They are a stronger positive correlation of percent protein with percent lysine, and lysine yield per hectare with 1000 grain weight, in o_2 maize than in normal maize.

The results presented here also show that there is more heterotic dilution in the o_2 hybrids than in the normal analogues for traits like percent protein, percent lysine (whole kernel basis) and percent lysine in protein. It has been observed that the inbred lines with the o_2 gene in recessive homozygous condition and a superior protein quality tend to produce hybrids of superior quality.

Maize (*Zea mays* L.) proteins have long been known as nutritionally poor, being deficient in the essential amino acids, lysine and tryptophan (Osborne, 1897, 1914 and Osborne and Mendel, 1914). It was only in 1964 that an increase in lysine content of the endosperm proteins by genetic means was demonstrated (Mertz *et al.*, 1964). It was shown that opaque-2 (o_2), a single recessive mutant of maize, was capable of increasing lysine by 69% in the endosperm proteins. Little is known yet of the background effects that influence this gene. Gupta and Kovács (1974a, b and c) have earlier compared diallel crosses of opaque-2 and analogous normal maize for a number of morphological features, yield and several of the yield components, and obtained interesting differences between the o_2 and the normal types. This present paper provides results on a number of quality traits in these crosses, with and without the presence of the o_2 gene in recessive homozygous condition, intercharacter relationships of these traits, their relationships with yield and a number of yield components related to kernel characteristics, and heterosis observed for these traits.

Materials and Methods

The experiment uses the same materials, six o_2 converted inbred lines and their normal analogues, and the 15 single cross hybrids obtained by diallel crossing of inbreds in each group (Gupta and Kovács, 1974b). Kernels obtained from a single sib-mated ear at the end of each row in the trial conducted during 1972 at Martonvásár were used for biochemical analyses. Kernels thus obtained from different replications for a given genotype were mixed together before obtaining samples for these analyses. Mean observations on kernel length, kernel width, 1000 grain weight, kernel density, percent water imbibing capacity of the kernels in 24 hours and grain yield expressed at 15% moisture level, all at normal population density (40,000 plants/ha), have been used in the presentation of this paper from Gupta and Kovács (1974a and c).

Crude protein (N × 6.25) was analysed in the samples using the boric acid modification of the Kjeldahl method (AACC, 1962). All the amino acids except those containing sulfur were analysed chromatographically, using the procedure developed by Moore and Stein (1954), in the nonoxidized hydrolysate obtained by the procedure described by Weidner and Eggum (1966). In their second oxidized hydrolysate, methionine was determined as methionine sulfone, and cysteine and cystine together as cysteic acid, which are the stable forms. Tryptophan, being acid labile, was unobtainable in both the hydrolyses. Oil was determined in the samples by the Soxhlet procedure (MSz, 1972).

The yield of protein, lysine (whole kernel basis) and oil per hectare were then calculated. Correlation coefficients were determined for a number of protein quality and oil traits, with each other and with the yield and yield components, by the method of Snedecor (1956), for the o_2 and normal types individually. Heterosis was calculated for each of the o_2 and normal single cross hybrids as the increase expressed as a percentage of their mid-parent (average of the two parents) values.

Table 1. *Observations on percent protein, protein yield, percent lysine, lysine yield, lysine in protein, percent oil and oil yield of the various o_2 and normal genotypes individually*

S. No. Genotype	% protein		Protein yield (kg/ha)		% lysine		Lysine yield (kg/ha)		g L/100 g P		% oil		Oil yield (kg/ha)	
	o_2	Normal	o_2	Normal	o_2	Normal	o_2	Normal	o_2	Normal	o_2	Normal	o_2	Normal
1. WF9	10.50	11.55	162.46	64.91	0.53	0.30	8.27	1.67	5.09	2.58	3.44	2.68	53.22	15.06
2. R61	11.55	12.60	465.23	189.05	0.53	0.34	21.22	5.07	4.56	2.68	2.94	2.45	118.42	36.76
3. N6	17.15	11.90	90.83	62.88	0.79	0.33	4.18	1.75	4.60	2.78	2.80	4.48	14.83	23.67
4. HMv850-2	10.85	10.15	310.96	191.71	0.50	0.29	14.40	5.50	4.63	2.87	3.09	2.75	88.56	51.94
5. C103	10.85	12.25	89.32	61.25	0.55	0.30	4.53	1.52	5.07	2.48	3.86	4.14	31.78	20.70
6. W187	15.05	11.90	35.04	66.12	0.71	0.35	1.65	1.92	4.70	2.91	2.54	3.97	5.91	22.06
7. WF9 × R61	8.75	12.25	405.61	654.84	0.36	0.31	16.92	16.70	4.17	2.55	2.63	3.51	121.92	187.63
8. WF9 × N6	11.20	12.25	375.78	618.43	0.42	0.33	13.94	16.51	3.71	2.67	3.03	3.51	101.66	177.20
9. WF9 × HMv850-2	9.45	10.15	446.46	419.56	0.40	0.30	18.97	12.50	4.25	2.98	3.15	2.29	148.82	94.66
10. WF9 × C103	9.45	10.50	370.86	514.50	0.37	0.35	14.46	16.92	3.90	3.29	3.47	2.05	136.18	100.45
11. WF9 × W187	9.80	11.90	252.64	339.20	0.42	0.35	10.84	9.84	4.29	2.90	3.47	3.73	89.46	106.32
12. R61 × N6	9.80	10.85	397.45	423.15	0.47	0.29	18.96	11.34	4.77	2.68	3.15	3.71	127.75	144.69
13. R61 × HMv850-2	9.80	9.10	592.70	508.84	0.39	0.25	23.47	14.15	3.96	2.78	3.63	4.35	219.54	243.23
14. R61 × C103	10.85	9.45	422.46	384.50	0.45	0.25	17.40	10.19	4.12	2.65	2.91	3.72	113.30	151.36
15. R61 × W187	9.10	10.15	387.44	518.71	0.36	0.28	15.38	14.52	3.97	2.80	3.13	3.75	133.26	191.64
16. N6 × HMv850-2	10.50	10.50	469.69	470.02	0.43	0.33	19.26	14.62	4.10	3.11	3.82	3.03	170.88	135.63
17. N6 × C103	11.55	11.90	419.77	552.02	0.36	0.28	12.93	13.08	3.08	2.37	3.37	4.28	122.48	198.54
18. N6 × W187	10.50	11.55	372.46	322.75	0.47	0.31	16.72	8.75	4.49	2.71	3.82	3.54	135.50	98.92
19. HMv850-2 × C103	8.40	10.15	420.30	518.58	0.35	0.29	17.53	14.83	4.17	2.86	2.89	3.03	144.60	154.81
20. HMv850-2 × W187	10.15	10.50	710.66	594.93	0.42	0.27	29.13	15.24	4.10	2.65	3.64	3.07	254.86	173.95
21. C103 × W187	9.80	11.55	413.52	484.73	0.39	0.28	16.38	11.83	3.96	2.44	3.12	4.05	131.65	169.97
Mean of parents	12.66	11.72	192.31	105.99	0.60	0.32	9.04	2.90	4.77	2,72	3.11	3.41	52.12	28.36
Mean of hybrids	9.94	10.85	430.52	488.32	0.40	0.30	17.49	13.40	4.07	2.76	3.28	3.44	143.46	155.27
Overall mean	10.72	11.10	362.46	379.08	0.46	0.30	15.07	10.40	4.27	2.75	3.23	3.43	117.36	119.01

Results

Observations on the protein quality and oil traits for the o_2 and the normal inbred lines and hybrids have been presented in Table 1. It will be seen that on average there is 10.72% protein in the o_2 genotypes compared with 11.10% in the normal types. If we express the o_2 as percent of normal, it is found to be about 96.6%, indicating an average difference of 3.4% in favour of the normal gene. The means of the parental inbred lines show that the o_2 lines have 12.66% average protein as against 11.72% in the analogous normal lines. Thus, the o_2 inbred lines have more protein in their kernels than their normal analogues. The o_2 single cross hybrids on the other hand show less protein, averaging 9.94%, than their normal counterparts which average 10.85%. The percent protein varies between 8.40 and 17.15 for the various genotypes in the presence of the o_2 gene in recessive homozygous condition and between 9.10 and 12.60 in the absence of the o_2 gene. It can be seen in Table 1 that the o_2 types yield an average of 362.46 kg of protein per hectare compared with the slightly higher average, 379.08 kg/ha, of the normal types, showing a 4.38% difference in favour of the absence of the o_2 gene. The parental inbred lines have averages of 192.31 and 105.99 kg protein/ha for the o_2 and normal types, respectively. The single cross hybrids average 430.52 and 488.32 kg protein/ha, respectively, in the presence and absence of the o_2 gene. The protein yield ranges between a minimum of 35.04 kg/ha and a maximum of 710.66 kg/ha for the o_2 inbred lines and hybrids and 61.25 and 654.84 kg/ha for the normal types.

The averages of all the genotypes for the trait percent lysine (whole kernel basis) are 0.46 and 0.30 for the o_2 and normal types, respectively (Table 1), demonstrating a 53.3% difference in favour of the o_2 recessive homozygotes. The averages of the o_2 and normal inbred lines for percent lysine are 0.60 and 0.32, respectively, showing a difference of 87%, and of the hybrids, 0.40 and 0.30, a difference of 33%. The lysine in the whole kernel ranges between 0.35 and 0.79, and 0.25 and 0.35%, for the o_2 and normal genotypes, respectively, including the parental inbred lines and the hybrids. The yield of lysine averages 15.07 and 10.40 kg/ha for the o_2 and normal analogues, respectively. The o_2 recessive homozygotes, thus, yield on average 44.9% more lysine per hectare than their normal analogues. The inbred lines average 9.04 and 2.90 kg lysine/ha for the two opacity factors, a difference of 211%, and the single cross hybrids 17.49 and 13.40 kg/ha, a difference of 30%. The lysine yield varies from 1.65 to 29.13 kg/ha for the o_2 inbred lines and hybrids and 1.52 to 16.92 kg/ha for the normal analogues. The observations on g lysine/100 g protein (g L/100 g P) show averages of 4.27 and 2.75 for the o_2 and normal analogous genotypes (Table 1). There is, thus, an average increase of 55.3% in lysine of protein in the 21 genotypes which is attributable to the presence of the o_2 gene in recessive homozygous condition. The inbred lines average 4.77 and 2.72 g L/100 g P, respectively, for the o_2 and normal types, showing a difference of 75%, whereas the single cross hybrids have 4.07 and 2.76, a difference of 47%. The ranges observed are 3.08 to 5.09 for the o_2 inbred lines and hybrids and 2.37 to 3.29 for the normal inbred lines and hybrids.

The oil content of the o_2 and analogous normal maize kernels averages 3.23 and 3.43%, respectively (Table 1), indicating the disadvantage of the o_2 gene to be nearly 5.83% in terms of the normal types. The parental inbred lines have averages of 3.11 and 3.41% oil in the presence and absence of the o_2 gene, respectively, the single cross hybrids 3.28 and 3.44%. The o_2 genotypes range from 2.54 to 3.86% for oil content and the normal types from 2.05 to 4.48%. The oil yield of all the o_2 genotypes shows average of 117.36 kg oil/ha and that of the normal genotypes 119.01 kg, only slightly higher than that of the o_2 types. The inbred lines average 52.12 and 28.36 kg of oil per hectare for the o_2 and normal types, respectively, whereas the single cross hybrids show averages of 143.46 and 155.27 kg/ha, respectively.

Observations on the whole amino acid profile of all the 21 genotypes, with and without the o_2 gene in recessive homozygous condition, are presented in Table 2, except for lysine which appears above in Table 1. It is interesting to note that the presence of the o_2 gene generally increases the amount of a number of amino acids compared with their normal analogues: aspartic acid increases by 51%, arginine by 45%, glycine by 35%, threonine by 13%, valine by 11%, histidine by 9%, and cystine and isoleucine only slightly. The rest of the amino acids are reduced by up to 10% under the influence of the o_2 gene in the inbred lines and their hybrids, except for leucine and methionine which are, on average, reduced by 28 and 15%, respectively, compared with their normal counterparts. There are genotypes, however, which show an increase in the content of these amino acids in the presence of the o_2 gene in recessive homozygous condition.

Correlation Coefficients

Inter-character correlation was studied among a number of quality traits: percent protein, percent lysine (whole kernel basis), percent lysine in protein (g L/100 g P) and percent oil, individually for the o_2 recessive homozygotes and the normal analogues. The difference between the two, o_2 and normal, analogous correlation coefficients (r) was also examined statistically. The results of these correlation studies are presented in Table 3. It will be seen that only the traits percent protein and percent lysine (whole kernel basis) show a difference in their r which is statistically significant, demonstrating that the two traits are more closely related to each other in the o_2 recessive homozygotes than in the normal analogues. The relationships between other sets of traits do not show any mathematical difference under the influence of the o_2 gene (Table 3). It may, however, be added that the trait, percent oil, tends to have an inverse relationship with other traits under analysis. Table 4 presents observations on the correlation coefficients studied between the various protein quality or oil traits on the one hand and yield or yield components related to kernel characteristics on the other. It is interesting to note that the traits, percent protein and percent lysine, tend to show an inverse relationship with yield and yield components except with the water imbibition capacity of the kernels in 24 hours, both in the presence and absence of the o_2 gene in recessive homozygous condition; this is mostly statistically significant, especially in the presence of the o_2 gene in recessive homozygous condition. On the other hand, the traits, protein yield and lysine yield, show a positive correlation with the yield and yield components except the water imbibition capacity, which is again statistically significant especially in the o_2 recessive homozygotes (Table 4). The percent protein in lysine (g L/100 g P) is significantly negatively correlated with grain yield, kernel length, and width in the o_2 recessive homozygotes but not in the normal analogues and also not with the other yield components. The percent oil does not demonstrate any significant correlation with the yield or yield components, whether negative or positive, except with 1000 grain weight in the normal genotypes where a significant positive correlation is visible. Oil yield behaves in the same manner as the yield of protein or lysine.

It will further be seen in Table 4 that the correlation coefficients calculated between the o_2 and the normal genotypes for a given character are statistically positively significant only in the cases of protein yield, lysine yield and oil yield. The traits, percent protein, percent lysine and percent lysine in protein, demonstrate a positive but non-significant correlation between the o_2 and the normal types. Percent oil on the other hand shows a non-significant but negative correlation between the o_2 and the normal types.

It must be stressed that the difference between the two analogous correlation coefficients, one at the o_2 level and the other at the normal level, for a given set of characters are statistically significant only in six cases out of a total of 42 comparisons (Table 4). In all except one case, to be mentioned later, the quality traits, such as percent protein with 1000 grain weight, percent lysine (whole kernel basis) with kernel length as well as 1000 grain weight, and percent lysine in protein with grain yield and also with kernel length, are more strongly inversely correlated

Table 2. *Observations on the whole amino acid profile except lysine*

S. No.	Genotype		Histidine	Arginine	Aspar. acid	Threonine	Serine	Glutamic acid	Proline
1.	WF9	o_2	3.06	6.84	10.91	4.17	4.58	16.46	7.80
		+	3.00	4.20	6.48	3.56	4.55	19.92	10.34
2.	R61	o_2	3.10	5.74	11.13	3.84	4.61	17.36	8.46
		+	2.17	3.83	7.93	3.61	5.03	20.56	9.83
3.	N6	o_2	2.62	6.02	12.27	4.63	4.80	15.96	6.99
		+	3.32	4.76	7.16	3.47	4.61	19.20	10.40
4.	HMv850-2	o_2	3.18	6.06	10.53	3.71	4.43	16.27	8.66
		+	3.11	4.52	7.41	3.47	4.56	18.39	9.30
5.	C103	o_2	3.24	6.95	12.10	3.88	4.13	15.48	8.26
		+	2.22	3.53	8.19	3.13	4.46	20.76	8.90
6.	W187	o_2	2.70	5.94	13.28	3.81	4.49	16.41	6.67
		+	2.80	4.69	7.27	3.47	4.65	18.81	8.90
7.	WF9 × R61	o_2	3.30	6.43	10.65	4.31	4.65	16.34	8.19
		+	2.63	4.09	6.43	3.38	4.34	17.61	8.45
8.	WF9 × N6	o_2	2.87	5.74	11.45	4.36	4.47	16.01	8.45
		+	3.03	3.74	6.82	3.22	4.41	19.75	10.70
9.	WF9 × HMv850-2	o_2	3.19	5.61	10.18	3.53	4.21	17.44	8.99
		+	3.20	4.66	7.08	3.32	4.21	18.53	9.35
10.	WF9 × C103	o_2	3.59	6.50	8.56	3.68	4.19	16.38	8.82
		+	3.36	3.66	6.20	3.26	4.40	19.82	10.80
11.	WF9 × W187	o_2	3.29	5.84	10.60	3.85	4.43	17.05	9.22
		+	2.93	4.04	6.42	3.38	4.42	19.96	9.28
12.	R61 × N6	o_2	3.21	6.01	12.05	3.96	4.38	15.80	8.86
		+	2.84	4.13	7.09	3.55	4.78	18.87	9.64
13.	R61 × HMv850-2	o_2	2.92	5.72	9.30	3.58	4.32	17.77	9.05
		+	2.75	4.18	6.73	3.44	4.47	19.30	9.53
14.	R61 × C103	o_2	2.97	5.39	11.36	3.75	4.05	15.95	9.22
		+	2.69	4.25	6.88	3.58	4.50	19.14	10.17
15.	R61 × W187	o_2	2.95	5.27	10.26	3.83	4.50	17.15	8.70
		+	2.49	4.05	7.30	3.64	4.95	20.05	9.52
16.	N6 × HMv850-2	o_2	2.55	5.58	10.98	3.97	4.14	16.83	8.65
		+	2.88	4.22	8.29	3.61	4.34	18.60	9.86
17.	N6 × C103	o_2	2.76	4.96	9.23	3.90	4.52	18.10	9.08
		+	2.26	3.19	7.04	3.45	4.68	21.13	9.32
18.	N6 × W187	o_2	3.08	5.47	10.95	3.92	4.52	16.26	9.01
		+	2.63	3.79	7.33	3.51	4.61	20.28	9.44
19.	HMv850-2 × C103	o_2	3.17	5.77	10.25	3.67	4.33	16.62	10.14
		+	2.74	4.02	7.72	3.54	4.57	19.23	10.20
20.	HMv850-2 × W187	o_2	2.96	5.73	9.66	3.66	4.43	17.02	9.08
		+	2.65	3.91	7.28	3.43	4.58	19.64	9.68
21.	C103 × W187	o_2	3.03	5.18	9.71	3.89	4.52	16.87	9.66
		+	2.58	3.44	6.25	3.32	4.50	20.05	10.04
	Mean	o_2	3.04	5.85	10.73	3.90	4.41	16.64	8.66
		+	2.78	4.04	7.11	3.44	4.55	19.50	9.70
	O_2 as % of normal		109.35	144.80	150.91	113.37	96.92	85.33	89.28

with the yield and yield components in the o_2 recessive homozygotes than in the normal analogues. The percent oil and oil yield do not show any significant difference between the o_2 and the normal analogous correlation coefficients with the yield and yield components. The only correlation coefficient of some interest to opaque-2 maize breeders is that of lysine yield per hectare and 1000 grain weight, which shows a r equal to 0.79*** at the o_2 level as against 0.07 for the analogous normal genotypes. The difference between the two r values is significant at a 1% level of significance.

Heterosis

Observations on the percent heterosis over the mid-parent have been presented in Table 5 for each of the o_2 and the normal single cross hybrids for the various protein quality and oil traits. It will be seen that percent protein demonstrates, in general, a negative heterosis over the mid-parent which is −20.7% in the case of the o_2 hybrids and −7.4% for the analogous normal hybrids. Thus, the o_2 hybrids have a protein content which is nearly 3 times lower than that of the analogous normal hybrids. Among the normal hybrids there are three which show a positive *of the various o_2 and normal genotypes (g amino acid/16 g N)*

Glycine	Alanine	Cystine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Fenyl-alanine	Ammonia
5.07	6.47	2.27	5.25	1.96	3.09	7.76	3.72	3.85	1.64
3.48	7.17	2.58	4.43	2.42	2.93	11.70	4.21	4.73	1.71
4.62	6.47	1.99	4.96	1.91	3.03	8.48	3.90	4.18	1.67
3.63	7.86	1.93	3.76	2.88	2.24	11.87	3.54	4.83	1.83
4.94	7.03	1.90	5.07	1.60	3.42	8.15	3.61	4.52	1.89
3.24	7.06	1.87	4.66	1.77	3.34	11.94	3.81	4.90	1.71
4.32	6.39	2.26	4.95	2.66	3.25	8.92	3.65	4.42	1.68
3.47	7.29	2.10	4.76	2.89	3.24	11.92	4.12	4.89	1.70
4.44	6.30	2.20	4.89	1.88	3.36	8.08	3.68	4.26	1.81
2.99	7.66	1.51	4.50	1.54	3.66	13.15	4.01	5.38	1.94
4.77	6.89	1.61	4.99	1.31	3.20	8.65	3.81	4.66	2.08
3.66	7.26	1.92	4.64	2.28	3.34	11.90	4.17	5.00	1.57
4.72	6.35	2.22	5.28	1.95	3.27	8.65	3.99	3.98	1.54
3.38	6.74	1.93	4.50	2.17	3.08	11.15	4.08	4.91	1.50
5.03	6.75	1.93	5.65	1.68	3.59	8.95	3.68	3.97	1.73
3.03	7.14	1.93	4.66	1.91	3.80	14.14	3.51	3.57	1.98
4.36	6.40	2.06	5.17	1.92	3.67	10.44	2.99	3.66	1.92
3.31	6.76	2.26	4.65	2.50	3.72	12.72	4.17	4.80	1.77
4.08	6.25	2.14	5.00	1.93	3.74	10.49	3.97	4.93	1.84
3.16	7.02	2.51	4.74	2.37	3.11	13.96	2.80	3.49	2.06
4.65	6.56	2.30	5.12	2.00	3.01	8.48	3.76	3.87	1.71
3.19	7.21	2.16	4.72	2.46	3.73	13.99	3.29	4.03	1.89
4.70	6.54	2.06	5.27	2.05	3.12	7.88	3.72	3.90	1.73
3.54	7.29	1.85	4.83	2.39	3.30	12.00	4.42	5.27	1.52
4.34	6.51	2.20	5.53	2.27	3.42	9.48	3.57	3.93	2.11
3.62	7.22	2.04	5.42	2.68	3.43	12.05	4.01	4.20	2.14
4.57	6.68	1.95	5.62	1.88	3.49	8.93	3.73	4.11	2.22
3.57	7.07	2.12	5.24	2.34	3.38	11.85	3.93	4.73	1.90
4.69	6.44	2.20	5.28	2.18	3.45	9.14	3.71	4.13	1.85
3.78	7.21	2.03	4.43	2.30	3.22	11.89	4.00	4.49	1.84
4.74	6.73	1.91	5.22	1.91	3.55	9.36	3.79	4.25	1.74
3.84	7.07	2.07	4.91	2.39	3.47	11.30	3.92	4.26	1.86
4.31	7.00	1.82	5.18	1.77	3.55	10.60	3.84	4.40	1.91
2.96	7.36	1.69	4.58	1.83	3.55	13.53	4.01	5.15	1.87
4.66	6.55	1.91	5.33	1.91	3.52	8.89	3.62	4.16	1.76
3.51	7.15	1.83	4.62	1.97	3.46	12.40	3.87	4.40	2.48
4.46	6.38	2.54	5.08	1.92	3.26	8.78	3.56	3.90	2.00
3.54	6.98	2.21	4.80	2.37	3.35	11.77	3.83	4.28	2.01
4.55	6.68	2.15	5.04	2.10	3.42	9.33	3.70	4.59	1.79
3.63	7.31	2.08	4.73	2.41	3.36	12.16	3.92	4.66	1.90
4.73	6.79	1.99	5.77	1.90	3.49	8.99	3.66	3.92	1.94
3.21	7.37	2.06	5.29	2.09	3.51	12.87	3.93	4.86	1.95
4.61	6.58	2.08	5.22	1.94	3.38	8.97	3.70	4.17	1.84
3.42	7.20	2.03	4.71	2.28	3.34	12.39	3.88	4.61	1.86
134.79	91.39	102.46	110.83	85.09	101.20	72.40	95.36	90.46	98.92

heterosis for percent protein, whereas among the o_2 hybrids there are none. The heterosis for percent protein ranges between −34.8 and 3.1% for the o_2 hybrids and between −23.9 and 4.5% for the normal hybrids. Table 5 shows that heterosis for protein yield in the o_2 hybrids is 189.7%, whereas the average for their normal analogues is 423.5%. The o_2 hybrids are, thus, less than half as heterotic as the normal hybrids for yield of protein per hectare. The ranges observed for the o_2 and the normal hybrids, respectively, are 29.2 to 56.5% and 167.3 to 789.4%.

The data on heterosis for percent lysine (Table 5) show the same trend as for percent protein. The o_2 hybrids show, in general, −32.5% heterosis and the normal analogues −6%. The heterosis in o_2 hybrids for percent lysine ranges between −46.9 and −17% and for normals between −21.9 and 14.8%. The o_2 hybrids further demonstrate 143.8% mean heterosis for lysine yield per hectare, whereas the normal analogous hybrids show 429.7%, nearly three times that of the o_2 hybrids. The o_2 hybrids vary in heterosis for lysine yield from 14.7 to 473.6% and the normal analogues from 167.7 to 960.8%. Percent lysine

Table 3. *Correlation coefficients observed between percent protein, percent lysine, g L/100 g P and percent oil at the o_2 and normal gene levels*[a]

S.No.	Character		% lysine	g L/100 g P	% oil
1.	% protein	o_2	0.90*	0.24	−0.32
		+	0.62	−0.41	0.17
2.	% lysine	o_2		0.64	−0.27
		+		0.46	−0.23
3.	gL/100g P	o_2			−0.01
		+			−0.52

[a] Correlation coefficients greater than 0.43, 0.54 and 0.66 are significant at 5%, 1% and 0.1% levels of significance, respectively. Degrees of freedom: 19.
* indicates significance of difference between the o_2 and the analogous normal correlation coefficients at 5% level of significance.

crosses involving a given inbred line have been presented in Table 6 individually for the o_2 and the normal types, in an attempt to study any possible relationship between the parental inbred lines and their single cross progeny and also the influence of the o_2 gene on this relationship.

It will be seen from the data presented in Tables 1 and 6 that the inbred line N6, which has a higher content of kernel proteins especially at the o_2 level, has a tendency to transmit a fairly high amount of protein to its single cross progeny, both at the o_2 and the normal levels. Similarly, the inbred line HMv 850-2, having somewhat smaller amount of kernel proteins both at the o_2 and the normal levels, tends to produce hybrids with a lower content of protein, both with and without the o_2 gene in recessive homozygous condition. A similar trend is visible for the trait, protein yield. The inbred line HMv850-2, having a higher yield of protein in the hybrid combinations than the other inbreds, both at the o_2 and the normal levels, has a fairly high yield of protein at the inbred level.

Table 4. *Correlation coefficients observed between the protein quality traits and yield and yield components*[a]

S. No.	Character		Grain yield	Kernel length	Kernel width	1000 grain weight	Kernel density	% water imbibition	O_2 and normal
1.	% protein	o_2	−0.68	−0.50	−0.70	−0.89*	−0.68	0.00	0.38
		+	−0.48	−0.33	−0.33	−0.62	−0.52	0.47	
2.	Protein yield	o_2	0.98	0.84	0.76	0.77	0.48	−0.30	0.77
		+	0.98	0.83	0.78	0.80	0.28	−0.66	
3.	% lysine	o_2	−0.77	−0.64**	−0.79	−0.86**	−0.51	0.53	0.39
		+	−0.39	−0.21	−0.47	−0.50	−0.38	0.43	
4.	Lysine yield	o_2	0.96	0.83	0.74	0.79**	0.55	−0.27	0.66
		+	0.98	0.83	0.74	0.07	0.28	−0.66	
5.	g L/100 g P	o_2	−0.53*	−0.55*	−0.53	−0.01	0.08	0.18	0.03
		+	0.09	0.12	−0.22	0.09	0.09	−0.05	
6.	% oil	o_2	0.21	0.03	0.18	0.20	0.30	−0.10	−0.05
		+	−0.07	−0.06	0.10	0.61	0.03	0.12	
7.	Oil yield	o_2	0.97	0.79	0.73	0.75	0.53	−0.26	0.73
		+	0.92	0.80	0.79	0.85	0.41	−0.65	

[a] Same as in Table 3.
*, ** indicate significance of difference between the o_2 and normal analogous correlation coefficients at 5% and 1% levels of significance respectively.

in protein demonstrates 14.6% negative heterosis at the o_2 level and 1.83% positive heterosis at the normal level, with ranges of −36.3 to 4.15% and −9.9 to 30% for the two levels of opacity, respectively (Table 5).

Table 5 also shows that percent oil shows 6.8% heterosis at the o_2 level and 2.8% at the normal level, ranging from −17.5 to 43.1% at the o_2 level and from −39.9 to 67.3% at the normal level. The oil yield shows 279% mean heterosis in the o_2 hybrids and 476% in the normal hybrids, with ranges from 42.1 to 1206.6% and 182.6 to 815.3% heterosis in the o_2 and normal hybrids, respectively.

Parent Progeny Relationships

In a six-parent diallel of the type used in the present studies each inbred line is involved in five single cross combinnations. Averages of all the 5 single

Percent lysine (whole kernel basis) shows that the inbred line N6, having a higher amount of lysine in its single cross hybrids, is itself relatively superior in its lysine content both at the o_2 and the normal levels. Similarly, the inbred lines HMv850-2 and R61, having relatively more lysine per hectare, have hybrids with a higher yield of lysine both at the o_2 and the normal levels. The transmission behaviour of the traits, percent lysine in protein and percent oil in the kernel, to the progeny is not clear in these studies either at the o_2 or the normal gene levels.

The yield of oil per hectare behaves in the same way as the yield of protein and lysine, that is, the inbred line HMv850-2 tends to be the higher yielder of oil both at the inbred and the hybrid levels and in the presence or absence of the o_2 gene in recessive homozygous condition.

Discussion

Undoubtedly, the effects of the o_2 gene in recessive homozygous condition are not underestimated in a comparison of crosses of the type used in the present studies (Gupta and Kovács, 1974a). The observations presented above have shown that the hybrids have a lower average for the characters, such as percent protein, percent lysine (whole kernel basis) and percent lysine in protein, than their parental inbred lines. This has been termed "heterotic dilution" and has long been known to occur with total protein in maize kernels (Hayes, 1922 and Frey, 1951). A point of disadvantage to the opaque-2 maize breeder observed in these findings is that this phenomenon of heterotic dilution is more prominent in the o_2 maize for all the three traits mentioned above than in the normal counterparts.

There is a nearly 30% increase in the harvest of lysine per hectare with the o_2 maize hybrids compared with the normal analogues, but the yields of total protein and oil per hectare are simultaneously diminished by nearly 12 and 8%, respectively.

Our results, like those of Dudley *et al.* (1971), have shown a significant negative correlation between percent protein and grain yield, and percent protein and kernel density, and a significant positive correlation between percent lysine and g L/100 g P, and percent lysine and percent protein. The observation that percent protein has a stronger correlation with percent lysine in the whole kernel in the o_2 recessive homozygotes than in the normal analogues corroborates that made by Paez and Zuber (1973) and should make it easier to select for higher percent protein with a simultaneous increase in the percent lysine in the whole kernel in the opaque-2 maize genotypes. Sreeramulu and Bauman (1970), on the other hand, observed a low but positive correlation between percent protein and grain yield both at the o_2 and the normal gene levels. The observation that there is a better correlation between the lysine yield per hectare and the 1000-grain weight in the opaque-2 maize could again be utilized advantageously by o_2 maize breeders.

The observation that the inbred lines superior in protein quality traits tend to produce hybrids of superior quality suggests that a selection of inbred lines to be used in breeding programmes is possible and desirable at the o_2 gene level.

Table 5. *Percent heterosis observed over mid-parent values for the various protein quality and oil traits for the o_2 and normal hybrids*

S. No.	Hybrid	% protein		Protein yield		% lysine		Lysine yield		g L/100 g P		% oil		Oil yield	
		O_2	Normal	O_2	Normal	O_2	Normal	O_2	Normal	O_2	Normal	O_2	Normal	O_2	Normal
1.	WF9 × R61	−20.63	1.45	29.24	415.70	−31.21	−1.70	14.75	395.55	−13.58	−3.04	−17.55	37.11	42.06	624.16
2.	WF9 × N6	−18.99	4.48	196.72	867.88	−37.20	4.04	123.93	865.50	−23.42	−0.37	− 2.88	− 1.96	198.82	815.29
3.	WF9 × HMv850-2	−11.47	−6.45	88.61	226.99	−22.53	2.68	67.36	248.67	−12.55	9.36	− 3.37	−15.50	109.93	182.57
4.	WF9 × C103	−11.47	−11.76	194.59	715.63	−32.04	14.79	125.94	960.81	−23.23	30.04	− 4.93	−39.88	220.42	461.80
5.	WF9 × W187	−23.29	1.49	155.84	417.74	−32.28	7.14	118.55	448.19	−12.36	5.65	16.05	12.35	202.64	472.84
6.	R61 × N6	−31.71	−11.43	42.95	235.93	−28.93	−12.99	49.29	232.55	4.15	−1.83	9.76	6.74	91.76	378.95
7.	R61 × HMv850-2	−12.50	−20.00	52.72	167.28	−24.57	−19.55	31.78	167.74	−13.82	0.18	20.60	67.31	112.14	448.43
8.	R61 × C103	− 3.12	−23.94	52.36	207.23	−16.98	−21.92	35.14	209.26	−14.43	2.71	−14.41	13.07	50.87	426.84
9.	R61 × W187	−31.58	−17.14	54.89	306.56	−41.44	−16.90	34.50	315.45	−14.25	0.18	14.23	16.82	114.38	551.61
10.	N6 × HMv850-2	−25.00	− 4.76	133.80	269.24	−33.32	4.98	107.32	303.31	−11.16	10.09	29.93	−16.07	230.59	258.81
11.	N6 × C103	−17.50	−1.45	366.02	789.42	−46.87	−11.12	196.90	700.00	−36.30	−9.88	1.20	− 0.70	425.67	795.13
12.	N6 × W187	−34.78	−2.94	491.82	400.39	−36.99	− 7.53	473.58	376.84	− 3.44	−4.75	43.07	−16.11	1206.65	332.72
13.	HMv850-2 × C103	−22.58	− 9.37	110.00	310.01	−33.43	− 2.42	85.21	322.51	−14.02	6.91	−16.71	−11.92	140.32	326.24
14.	HMv850-2 × W187	−21.62	− 4.76	310.79	361.49	−31.20	−15.62	262.99	310.78	−12.11	−8.30	29.54	− 8.63	439.61	370.14
15.	C103 × W187	−24.32	− 4.35	565.04	661.14	−38.27	−13.29	430.10	587.79	−18.94	−9.46	− 2.50	0.00	598.78	695.00
Mean		−20.70	− 7.40	189.69	423.51	−32.48	− 5.96	143.82	429.66	−14.63	1.83	6.80	2.84	278.98	476.04

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Table 6. *Average performance of a given inbred line in hybrid combinations*

S. No.	Inbred line		% protein	Protein yield (kg/ha)	% lysine	Lysine yield (kg/ha)	g L/100 g P	% oil	Oil yield (kg/ha)
1.	WF9	o_2	9.73	370.27	0.39	15.03	4.06	3.15	119.61
		+	11.41	509.31	0.33	14.49	2.88	3.02	133.25
2.	R61	o_2	9.66	441.13	0.41	18.43	4.20	3.09	143.15
		+	10.36	498.01	0.28	13.38	2.69	3.81	183.71
3.	N6	o_2	10.71	407.03	0.43	16.36	4.03	3.44	131.65
		+	11.41	477.27	0.31	12.86	2.71	3.61	151.00
4.	HMv850-2	o_2	9.66	527.96	0.40	21.67	4.12	3.43	187.74
		+	10.08	502.39	0.29	14.27	2.88	3.15	160.46
5.	C103	o_2	10.01	409.38	0.31	15.74	3.85	3.15	129.64
		+	10.71	490.87	0.29	13.37	2.72	2.62	155.03
6.	W187	o_2	9.87	427.34	0.41	17.69	4.16	3.44	148.95
		+	11.13	452.06	0.30	12.04	2.70	3.63	148.16

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