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BREEDING FOR TOLERANCE TO IRON CHLOROSIS AND QUALITY CHARACTERS THROUGH ANTHER CULTURE IN RICE

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ABSTRACT

Anther culture was under taken in three hybrids of rice with an object to isolate genotype having tolerance to iron chlorosis and improved grain quality. Callus formation and plant generation was observed only in the hybrid Prabhavati × Basmati 370, which was also very low. The spontaneously diplodised plants were studied for their trueness. Their progenies were studied for the important characteristic and advanced. Variability was quite evident among the different progenies for the characters studied. Promising cultures were evaluated in multilocation trials for three years. One of the cultures Parag 401 has been released for cultivation under upland irrigated conditions. It is superior to the parent Prabhavati and check Sugandha for yield and quality characters having tolerance to iron chlorosis.

Key words: Anther culture, quality characters, rice

Pollen culture offers an efficient experimental system, for genetic manipulation. The good complementary characteristics of two parents can be combined in pollen plants and in one generation the dihaploid plants can be developed. After the initial reports of successful production of haploids from anther culture in Datura [1], haploids have been obtained in more than 300 species of angiosperms. Several workers emphasised the importance of anther culture in rice and reported studies on callus formation and plant regeneration [2 - 4]. From a cross of *indica* × Basmati rice, anther culture in F_1 hybrids resulted in isolation of genotypes having desirable agronomic characteristics [5]. Similarly a salt tolerant doubled haploid line through anther culture has also been reported [6].

The black cotton soils of Maharashtra are rich in organic matter and are calcareous in nature. Due to iron chlorosis, cultivation of new semidwarf varieties of rice in these areas is a major constraint. Similarly the presently grown varieties are having medium coarse grains. Rice varieties with fine grain and aroma are highly priced and liked by the consumers.

The present study was undertaken with a view to isolate the haplodiploid lines having good cooking quality characteristics and tolerance to iron chlorosis through compaction of early generations using the anther culture technique.

MATERIALS AND METHODS

The F_1 hybrids viz., the Prabhavati × Basmati 370, Prabhavati × IET8573 and Prabhavati × Karnal local were grown along with parents on the experimental farm of the department. The true hybrid plants were identified. The media used for anther culture was N6 [7] with inosital (1.0 g/1), sucrose (3%) and 2, 4-D (2mg/1). After inocculation of the anthers, cold treatment was given at 6°C for 24 hrs. Calli developed were sub cultured twice on the same medium. The proliferated calli were subsequently transferred in the medium containing 0.5 mg/l NAA and 1.0 mg/l Kinetin and kept for regeneration at 22 ± 1°C alternating the light and dark period of 18 and 6 hrs respectively. The rooting medium was supplemented with 2 mg/l NAA. The completely regenerated plantlets were transferred in the field.

The seeds of the surviving plants were sown during the next kharif season alongwith the parents to confirm the dihaploid nature. The diploid nature of the dihaploid selections was confirmed from non segregation (uniformity) of the characters. The selected progenies were subsequently studied for different characteristics including seed sterility. The selections were based on the number of tillers per plant, panicle length, grains per panicle and grain characteristics. Tolerance to iron chlorosis was one of the important criteria for selection.

The promising progenies were advanced during off season at the Directorate of Rice Research, Hyderabad and subsequently evaluated for variability in preliminary and multilocation trials.

RESULTS AND DISCUSSION

In the cross Prabhavati × Basmati 370, the pollen derived calli emerged through the split anther lobes of some of the anthers. However, the anther response was much lower (Table 1). The response to calli formation was only 1.2% in the cross Prabhavati × B 370 [8]. It was zero in other two hybrids viz., Prabhavati × IET 8573 and Prabhavati × Karnal local. In general *Indica* varieties are less responsive to anther culture than *japonica* types [9]. Similarly, the response in different hybrids and varieties of rice was also variable and low [10, 11]. In the present study the plant regeneration frequency from the callus was also very poor. Among the regenerants very high frequency of *albina* types was observed (Table 1). Some of the plants had reduced vigour and narrow leaves with pale green colour. These were probably

Table 1. Anther response for callus formation and pollen plant regeneration in crosses of rice

Cross	Particulars				
	Anthers inoculated	Callus formation	Shoot/root regeneration	Green plants	Albino plants
Prabhavati × B 370 (Lot No. 5)	560	7 (1.2%)	45	22	23
Prabhavati × IET 8573 (Lot No. 5)	450	Callus could not be obtained			
Prabhavati × Karnal local (Lot No. 5)	530	Callus could not be obtained			

haploids. The other normal plants might have spontaneously diplodised. The frequency of spontaneous doubling of chromosomes during the course of *in vitro* culture is reported to the extent of 80% [11]. Similar results were also reported by other scientists [12]. It was interesting to note that the regenerated plants had anthocynin pigmentation on husk on the seed. This anthocynin disappeared subsequently. Seed sterility ranged from 20 to 25%. Grains of these plants were longer than Basmati 370 and transluscent.

Variability analysis from anther culture derived lines showed shift of progeny mean in both the directions (Table 2). A wide range of variation was observed among progenies of the culture derived plants. The important characters like length of panicle, 1000 grain weight, number of grains per panicle, days to maturity and yield showed desirable shift of means the check Prabhavati. Genotypic coefficient of variability was high for grain yield per plot. However, the heritability estimate was low. The anther culture derived variability has been reported earlier in rice [13, 14]. Greater variation in the regenerated plants than the parental line Calrose 76 for the characters grain length, protein content and plant height has been reported [15].

Selected lines having field tolerance to iron chlorosis, good grain quality characteristics and yield were evaluated in multilocation trials for three seasons (Table 3). The results showed that the genotype ACR 401 is tolerant to iron chlorosis, having 12% increase in yield over the check Sugandha with good grain quality characters. This genotype gave 36.5 q/ha yield which was on par to one of the parents Prabhavati and check Sugandha. It was semidwarf, maturing in 100-115 days and resistant to iron chlorosis [16].

Table 2. Parameters of genetic variability for grain yield and yield contributing characters

Sr. No.	Characters	Range	Mean	± SE	PCV (%)	GCV (%)	Heritability (%)
1.	Plant height (cm)	50.0-97.5 (62.5)	67.35	3.88	12.86	9.96	59.90
2.	No. of effective tillers per meter length	62.0-118.5 (102.5)	89.64	8.12	19.77	15.06	57.99
3.	Length of panicle (cm)	18.1-25.8 (20.5)	22.26	1.06	8.60	5.28	37.70
4.	1000-grain weight (g)	14.3-25.0 (23.5)	19.87	0.81	13.45	12.15	81.63
5.	No. of grains per panicle	47.3-105.6 (66.0)	77.15	8.71	21.07	13.74	42.55
6.	Days to 50% flowering	74.5-100.5 (81.0)	88.26	0.62	5.86	5.77	97.04
7.	Days to maturity	105.0-136.0 (117.0)	114.68	0.46	4.82	4.79	98.59
8.	Grain yield per plot (g)	92.0-338.5 (272.5)	221.45	36.21	30.67	20.14	43.12

The values in parenthesis are the mean value of Prabhavati

Table 3. Performance of ACR 401 in advance yield trials

Sr. No.	Year	Culture	Locationwise grain yield (kg/ha)			Mean	% increase over check Sugandha	Overall mean of location
			Parbhani	Nagpur	Kopergaon			
1.	1993-94	ACR 401	3953	2058	3408	3140	8.00	
		Sugandha	3436	2036	3248	2907		
		S.E. ±	187.0	140.0	97.53			
2.	1994-95	ACR 401	3703	1480	1645	2276	15.39	
		Sugandha	3383	2098	1427	1969		
		S.E. ±	193.72	217.95	168.80			
3.	1995-96	ACR 401	3374	2080	1755	2403	14.32	2606
		Sugandha	3209	2003	2054	2102		2326
		S.E. ±	212.92	276.00	76.92			
Mean		ACR 401	3677	1873	2269			
		Sugandha	3342	1712	1923			

As regards the grain quality characters ACR 401 has aroma, the grains are long, slender having 4.02 L/B ratio and intermediate gelatinization temperature (Table 4). It is superior to Prabhavati the earlier released variety of rice and the parental

Table 4. Performance of anther culture rice lines and parents for quality characters

Genotype	Kernel (mm)		L/B ratio	Grain type	Kernel elongation after cooking	Geletinization temperature	Protein content (%)
	Length	Breadth					
Prabhavati	6.93	2.16	3.20	MS	2.275	L	7.0
Basmati 370	7.49	1.84	3.91	LS	2.000	I	8.4
ACR 401	7.85	1.95	4.02	LS	1.530	I	8.3
ACR 52	8.80	2.00	4.40	LS	1.570	I	8.2

LS = Long slender, MS = Medium slender, L = Low, I = Intermediate

line of cross. This ACR 401 genotype has been released as Parag 401 for the irrigated upland rice cultivation in vertisols.

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