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INDUCED MUTATION FOR TUNGRO RESISTANCE IN RICE

Tungro is the most serious virus disease of rice in South and Southeast Asia. It is a composite disease of two kinds of viruses, rice tungro bacilliform virus (RTBV) and rice tungro spherical virus (RTSV). Damage to the plant is mostly caused by RTBV, while RTSV acts to facilitate RTBV acquisition and transmission by insect vector. Both viruses are transmitted mainly by green leafhopper (GLH). Resistance to GLH is common in rice germplasm but extremely rare for the two viruses. To induce mutations for tungro resistance, a susceptible variety IR22 was treated with N-methyl-N-nitrosourea (MNH) following the procedure of Satoh and Omura [1]. The panicles of rice variety 'IR22' were soaked in 1 mM MNH solution for 45 minutes at 16 to 18 hours after flowering.

Two thousand six hundred and forty fertile M_1 plants were produced. From these plants M_2 lines with 10 or more seedlings were planted in the field to evaluate their reaction against tungro under natural conditions in the 1990 dry season on the IRRI central research farm, Los Banos, the Philippines. Of these, 124 M_2 lines were selected by visual evaluation. Five plants were harvested individually from each selected line. A bulk was also made from all the remaining plants in the line. In the M_3 generation, each family consisted of five sister lines and one bulked line. One line (M_3-723) showed no tungro symptoms and its related bulk segregated for resistance but all other M_3 lines from the same family were susceptible to tungro. The resistant line, M_3-723, showed low infection with RTBV and RTSV when leaves were tested by enzyme-linked immunosorbent assay (ELISA) to diagnose tungro infection. All M_4 lines from M_3-723 showed uniform resistance in the field. They were not infected with RTBV and were resistant to RTSV infection (Table 1). The reaction of these plants to the virus vector GLH was variable.

To investigate the resistance of the M_4 lines in the field, they were inoculated with viruliferous GLH at the 10-day-old seedling stage in the laboratory. The rate of tungro infection of these selections, at 14 days after inoculation, was as high as the susceptible variety IR22 (Table 1). When the lines were diagnosed at 30, 60 and 90 days after inoculation, the infection rate did not decrease with the sampling date (Table 1). This indicates that the mutant lines are susceptible to tungro infection at the seedling stage and the infected plants do not recover from the disease. To determine whether the resistant reaction in the mutants differ among the growth stage, the progeny of M_4 lines, that showed no infection with either RTBV or RTSV in the field but showed low level of antibiosis to GLH, were used. They were inoculated with viruliferous GLH at 10, 24, 38, 52, 66 and 88 days after seeding, respectively. Both RTBV and RTSV infection rate decreased rapidly with age in selected M_5 lines (Table 2). Those mutant lines slightly susceptible to RTSV at the seedling stage were resistant at early tillering stage 24 days after seeding. All the mutant lines were susceptible to RTBV at the young seedling stage but became resistant at maximum tillering stage (52-66 days after seeding). It was therefore concluded that these mutant lines have resistance to both RTBV and RTSV infection at the maximum tillering stage, and they possess adult plant resistance to tungro.

Table 1. Reaction of mutant lines resistant to tungro in the field and in the laboratory on different sampling stage

M_4 line and variety	Plant [1] derived from M_3	Infection in field (%)			Infection in laboratory when 10-day-old seedlings were tested (%)							
		Plants tested (No.)	At maturity[2]		Plants tested (No.)	14 DAI[3]		30 DAI		60 DAI		90 DAI	
			RTBV	RTSV		RTBV	RTSV	RTBV	RTSV	RTBV	RTSV	RTBV	RTSV
453	723-1	24	5	19	25	92	4	92	4	92	8	92	8
454	723-2	11	0	9	3	100	0	100	0	100	0	100	0
455	723-3	18	0	11	26	89	4	92	15	96	15	96	15
456	723-4	19	0	26	24	83	0	83	12	88	17	88	12
457	723-5	20	0	10	12	92	0	92	8	92	0	92	0
458	723-6	23	0	17	9	78	11	89	22	100	22	89	22
459	723-7	18	0	11	9	89	0	100	0	100	0	100	0
460	723-8	22	0	23	14	100	0	100	21	100	29	100	21
461	723-9	24	0	25	13	92	0	100	8	100	8	100	8
IR22		24	92	92	16	94	19	100	81	-	-	100	100

[1] Line and plant number of M_3 generation; [2] Leaves were sampled at maturing stage in the field to diagnose for tungro infection, on the other hand, they were sampled on different days after inoculation in the laboratory; [3] DAI; days after inoculation

Table 2. Percentage of tungro infection in 20 plants each of M_5 lines on different inoculation dates for ELISA test

M_5 line and variety	Plant [1] derived from M_4	10 DAS[2]		24 DAS		38 DAS		52 DAS		66 DAS		80 DAS	
		RTBV	RTSV	RTBV	RTSV	RTBV	RTSV	RTBV	RTSV	RTBV	RTSV	RTBV	RTSV
H 6	460-6	100	5	40	0	55	0	25	0	5	0	5	0
H 10	460-10	100	40	45	0	30	0	20	0	15	5	5	0
I 7	461-7	95	5	50	0	30	0	35	0	5	0	0	0
I 9	461-9	90	10	55	0	45	0	30	0	5	0	0	0
IR22		95	45	100	55	100	25	100	55	50	0	65	0
TN1		100	90	80	70	80	50	100	25	50	15	35	10
ARC11554		15	5	5	0	0	0	0	0	0	0	0	0

[1] Line and plant number of M_4 generation; [2] DAS; days after seedling

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RICE MUTANTS OBTAINED THROUGH SODIUM AZIDE (NaN_3) TREATMENT

The successful utilization of sodium azide to generate genetic variability in plant breeding has been reported in barley [3], [4], rice [1], [2] and other crops. Rice seeds of 'Dourado Precoce', Brazilian upland cultivar, were treated with 5 x 10^{-3} M of sodium azide, prepared in buffer solution of pH 3,0, for 8 hours at laboratory temperature. Ten short culm mutant lines were selected in the M_2, M_3 and M_4 generations. They were denominated 1, 2, 3, 4, 5, 6, 8, 9, 10 and 13. In the M_5 generation, the mutant lines were evaluated for flowering and maturing cycles, tiller number per plant, plant height, panicle number per m^2, panicle length, fertility of panicle, weight of 1.000 grains, productivity, percentage of intact grains after milling, width and thickness of peeled and polished grains and length/width grain ratio. The experiment was conducted in the Centro Experimental of Instituto Agronômico, Campinas, São Paulo, Brazil, during the period of 1993/94, utilizing randomized block design with four replications. Each experimental plot consisted of five rows of four meters in length, 50 cm between rows, with 75 seeds sown per meter. The cultivar 'IAC 201' and the original Dourado Precoce were planted as checks. All observations were made on the three central rows of each experimental plot. The data was analysed by the SANEST statistical program and the mean values were discriminated by the Tukey's test at the level 5% of probability (Table 1).

Table 1. Mean values of various characters of 10 selected mutant lines, compared with original variety Dourado Percoce using Tukey's test

Mutant/lines	TNP	PH	PL	NFSP	PSSP	WTG	P	GW	GT	GLW R
1	2,16	124,64	20,50	80,84*	11,31	37,23*	5,587	2,53	2,02	3,00*
2	2,10	132,34	22,57	98,41	9,70	36,86	5,953	2,57*	2,05*	2,85
3	1,98	133,74	22,48	101,16	11,90	37,52*	6,433	2,56	2,03	2,84
4	2,53	106,60*	20,71	84,74*	7,47	31,16*	5,703	2,48	1,81*	2,79
5	2,75	107,70*	20,57	80,73*	6,17	30,53*	5,978	2,58	1,87*	2,76*
7	2,85*	112,55*	21,43	84,35*	4,49	31,16*	6,712	2,49	1,84*	2,79
8	2,81*	83,50*	16,75*	49,84*	20,05*	29,93*	2,833*	2,28*	1,95	3,04*
9	2,46	112,00*	20,13*	79,04*	4,69	30,00*	5,908	2,46	1,83*	2,78
10	2,56	107,60*	18,76*	68,42*	6,58	30,68*	5,545	2,48	1,83*	2,77*
13	2,16	128,25	20,66	89,20	11,61	37,28*	5,275	2,55	2,02	2,83
Dourado Precoce	1,99	135,84	23,00	114,84	6,70	34,36	6,278	2,48	1,96	2,86
IAC 201	2,25	119,94*	23,11	169,53*	6,58	24,21*	6,217	1,97*	1,69*	3,59*

*/ significant at the level of 5%
TNP = tiller number per plant
PH = planta height
WTG = weight of 1.000 grains
PL = panicle length
NFSP = number of fertile spikelets per panicle
PSSP = Percentage of sterile spikelets per panicle
P = Productivity
GW = grain width
GT = grain thickness
GLWR = grains length/width ratio