



This document has been produced and supplied by  
 The British Library Document Supply Centre, Boston Spa, West Yorkshire, UNITED KINGDOM, LS23 7BQ  
 WARNING: Further copying of this document, other than that allowed under the copyright law, is not permitted without the permission of the copyright owner or an authorised licensing body.

*2002  
 126002*

Table 2. Comparison of segregation for 8 heterozygous isozyme markers among F<sub>2</sub> plants, and anther culture-derived calli of the cross IRAT177/Apura.<sup>a</sup> IRII, 1988.

Materials	Locus							
	<i>Icd-1</i>	<i>Pgi-1</i>	<i>Sdh-1</i>	<i>Acp-1</i>	<i>Est-9</i>	<i>Amp-2</i>	<i>Pgd-1</i>	<i>Est-1</i>
IRAT177	F	F	F	F	S	S	F	A
Apura	S	S	S	S	F	F	S	P
F <sub>2</sub> progeny <sup>b</sup>	54:95:40	45:94:50	43:102:44	49:93:43	57:87:43	50:74:64	63:71:44	41:148
	ns	ns	ns	ns	ns	S**	S**	ns
AC-derived calli <sup>b</sup>	234:210	182:264	216:226	92:90	214:186	186:258	293:137	135:115
	ns	S**	ns	ns	ns	S**	S**	ns

<sup>a</sup>The allele designations adopted are F (fast) vs S (slow) when the parents differed in the migration speed of the allozymes, and A (absence) vs P (presence) when the parents differed in the presence of a band. <sup>b</sup>Segregation of isozyme phenotypes following the order SS:SF:FF or AA:AP:PP for the F<sub>2</sub> progeny and SS:FF or AA:PP for the AC-derived calli. By the *Chi Square Tests for Homogeneity*, F<sub>2</sub> fit a 1:2:1 or a 1:3 segregation; AC-derived calli fit a 1:1 segregation. S\*\* = significant at the 5% level, ns = not significant.

In the F<sub>1</sub>s involving distantly related rice varieties, a large number of heterozygous isozyme markers exist. Segregation of such markers can be surveyed in AC plants and in AC calli; calli are produced in larger numbers.

The isozyme genes listed in Table 1 have been found to be reliably expressed among microspore-derived calli. We utilized them as efficient markers in detecting *in vitro* selection occurring during the rice AC process. Table 2

displays segregation data of 8 isozyme markers among microspore calli and F<sub>2</sub> plants derived from a japonica × indica cross. In addition to the deviations in both F<sub>2</sub> plants and AC calli that are probably due to the hybrid breakdown that occurs in crosses between distantly related varieties in rice, the AC process itself seems to induce a slight deviation at the *Pgi-1* locus toward the japonica allele. This suggests that the hybrid

sterility breakdown that hampers the use of japonica × indica crosses in rice conventional breeding may also affect doubled haploid breeding. It also indicates that AC calli do not represent a fully random array of this modified gametic pool. Isozymes appear to be efficient markers for detecting *in vitro* selection occurring during the AC process, from the callus stage through the regenerated plant level. □

### Esterase isozyme as a marker in *in vitro* studies of rice

M. Maheswaran and S. R. S. Rangasamy, School of Genetics, Tamil Nadu Agricultural University, Coimbatore 641003, Tamil Nadu, India

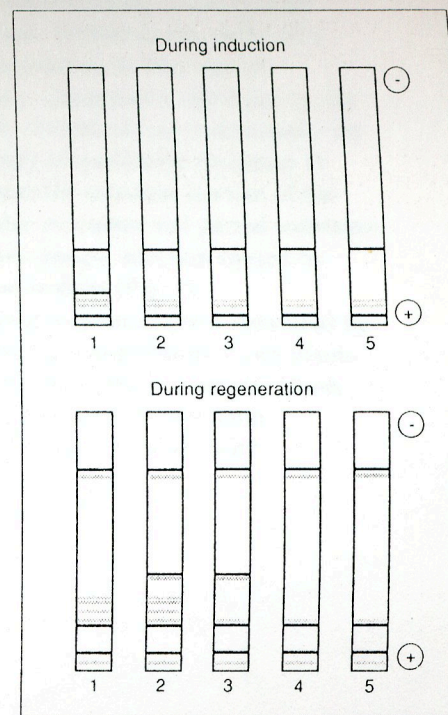
Isozymes can be used as markers during morphogenesis in *in vitro* studies. We studied the banding pattern of esterase isozymes in 21-d-old callus and regenerating callus of rice — *Oryza spontanea*, *O. glaberrima*, and *O. sativa* (cultivars Co 43, IR50, and IR1552).

The esterase enzyme was extracted using 0.2 M Tris-HCl buffer (pH 6.0) containing 0.006 M β-mercaptoethanol. The technique was vertical acrylamide gel electrophoresis using 8% acrylamide. The Tris (0.005 M) and glycine (0.038 M) mixture used as gel and electrode buffer had pH 8.3. Zymograms were revealed by staining with α-naphthyl acetate in acetone and fast blue RR salt mixture in sodium phosphate buffer (pH 6.2).

The similarities and dissimilarities in the zymograms of *O. spontanea*, *O. glaberrima*, and *O. sativa* show the species relationships of *Oryza*.

The electrophoresis of 21-d-old calli induced in Murashige and Skoog (MS) medium—each liter with 2 mg 2,4-dichlorophenoxy-acetic acid (2,4-D) and 0.5 mg kinetin—showed 3 common groups of fast-migrating bands in all 5 genotypes (see figure). An additional fast-migrating band observed in *O. spontanea* was absent in the other genotypes.

In the zymogram of regenerating calli in MS medium—with 1 mg kinetin and 1 mg naphthaleneacetic acid (NAA) per liter of the medium—*O. sativa* cultivars IR50 and IR1552 had similar banding patterns different from those of *O. spontanea* and *O. glaberrima*. *O. sativa* cultivar Co 43 had an additional band as well as the bands observed in IR50 and IR1552. *O. glaberrima* had an additional slow-migrating band not observed in *O. spontanea* but present in Co 43. Callus induction and plant



Zymogram of esterase isozymes of 5 *Oryza* genotypes, Tamil Nadu, India. 1 = *O. spontanea*, 2 = *O. glaberrima*, 3 = *O. sativa* Co 43, 4 = IR50, and 5 = IR1552.

Species and cultivar	Seeds inoculated (no.)	Seeds (no.) with callus	Callus production <sup>a</sup> (%)	Calli inoculated (no.)	Regenerating calli (no.)	Regeneration efficiency <sup>b</sup> (%)
<i>O. spontanea</i>	68	26	38.2	42	12	28.5
<i>O. glaberrima</i>	73	40	54.5	42	9	21.4
<i>O. sativa</i>						
Cultivar Co 43	62	15	24.6	24	2	4.5
Cultivar IR50	68	42	61.7	44	18	40.9
Cultivar IR1552	72	39	54.2	36	7	19.4

<sup>a</sup>Callus production (%) =  $\frac{\text{no. of seeds with callus}}{\text{no. of seeds inoculated}} \times 100$ . <sup>b</sup>Regeneration (%) =  $\frac{\text{no. of regenerating calli}}{\text{no. of calli inoculated}} \times 100$ .

regeneration frequencies are given in the table.

The banding patterns of esterase isozymes could be utilized as markers in *in vitro* studies by correlating banding patterns with frequencies of callus induction and regeneration. □

*The International Rice Research Newsletter* is published to expedite communication among scientists concerned with rice research and the development of improved technology for rice and rice-based farming systems. Readers are encouraged to write authors at their published addresses to discuss the research and obtain more details.

## Disease resistance

### Reaction of selected cultivars to tungro (RTV) and other diseases in Tamil Nadu

V. Mariappan, V. Narasimhan, M. Muthusamy, S. Muthusamy, and P. Vivekanandan, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Madurai 625104, India; and G. S. Khush, IRRI

All locally grown cultivars were severely damaged by RTV in 1984. We screened

newly selected breeding lines for resistance to RTV using the vector green leafhoppers for artificial inoculation at 10 d. Lines showing 0-10% infection were compared with popular local varieties ADT36, CO 37 (Vaigai), White Ponni, and IR20 in the field.

Six lines (IR32429-148-1-3-3, IR35366-90-3-2-1-2[IR72], IR37865-29-3-1-3, IR39357-91-3-2-3, TNAU831520, and TNAU831521) were least infected with leaf blast (0-2 grade), neck blast (1.6 to 12.8%), and brown spot (1-3 grade) and showed sheath rot moderate infection (3-5 grade). The four popular local varieties showed high RTV

infection with artificial inoculation but moderate resistance to other diseases in the field. The yield potential of the six cultures was comparable with that of the local varieties, or even better (see table). □

### A conceptual model of disease resistance in rice pathosystems, and its implications for evaluating resistance

S. W. Ahn and M. F. Koch, IRRI

We developed a conceptual model to focus understanding of quantitative resistance, defined as the ability of a host population to limit disease intensity. Quantitative resistance in rice usually consists of two components: the efficiency of qualitative resistance to eliminate the avirulent portion of the available inoculum and partial resistance to lower disease infection caused by virulent isolates (Fig. 1).

Partial resistance is best measured by challenging a population of rice plants with selected virulent isolate(s) alone, avoiding exogenous inoculum (alloinfection). A variety with

Yield and reaction to diseases of rice lines in Tamil Nadu, India.

Line or variety	Plants infected with RTV <sup>a</sup> (%)	Leaf blast <sup>b</sup> (grade)	Neck blast <sup>b</sup> (%)	Sheath rot <sup>b</sup> (grade)	Brown spot <sup>b</sup> (grade)	Mean yield (t/ha)
IR32429-148-1-3-3	0.0	2	3.3	3	3	5.7
IR35366-90-3-2-1-2 (IR72)	0.0	0	2.0	3	1	6.8
IR37865-29-3-1-3	10.0	0	12.8	5	2	5.1
IR39357-91-3-2-3	10.0	2	7.8	3	3	6.1
TNAU831520	6.3	0	1.4	5	3	5.9
TNAU831521	5.9	0	1.6	5	3	6.4
<i>Local checks</i>						
ADT36	70.0	0	19.4	5	3	5.1
CO 37	50.0	0	20.0	5	2	5.5
White Ponni	20.0	3	—	3	3	4.9
IR20	66.7	3	12.5	5	2	5.2

<sup>a</sup>Artificial inoculation. <sup>b</sup>Field infection under natural conditions.