

Exploitation of Twoline Heterosis Utilizing Thermosensitive Genic Male Sterility System in Rice (*Oryza sativa* L.)

Sasikala R^{1*}, R. Kalaiyarasi² and M. Paramathama³

ABSTRACT: Twoline hybrid rice breeding exploiting innovative tool viz., thermosensitive genic male sterile (TGMS) system has great potential in tropical countries like India. Hence an investigation was carried out by using four TGMS viz., TNAU 27 S, TS 09 12, TS 09 15 and TS 09 25 and fifteen pollen parents by utilizing Line x Tester design. Totally sixty hybrids and nineteen parents were raised along with two checks CORH 3 and Improved white ponni in Randomized Block Design with two replications. The F₁ twoline hybrids were evaluated for mean performance, gca and sca effects and heterosis for yield and yield components. Pre-ponderance of non-additive gene action for yield and yield attributing traits was observed by analyzing combining ability. TGMS lines TNAU 27 S and TS09 25 and the testers KDML 105, CB 05 911/884, BPT 5204 and CB 05/501 were good general combiners. The twoline hybrids combinations viz., TNAU 27 S x KDML 105, TS 09 25 x KDML 105, TNAU 27 S x CB 05 911/884, TNAU 27 S x BPT 5204 and TNAU 27 S x CB 05/501 exhibited good per se performance, high sca and high standard heterosis for yield over checks and the these hybrids could be utilized for commercial exploitation of twoline hybrids in rice. The present study revealed that the TGMS system may be utilized for developing high yielding twoline rice hybrids in India.

Key words: GCA & SCA variance, per se performance, gca and sca effects, heterosis

INTRODUCTION

The discovery of the environment sensitive genic male sterility (EGMS) system laid the foundation for re-placing three line system with the simpler and more efficient two-line system for hybrid rice seed production. EGMS includes photo period – sensitive genic male sterility (PGMS) and thermosensitive genic male sterility (TGMS) system. The discovery of Nongken 58 S in 1973, a PGMS/TGMS japonica rice line [1], provided the first genetic source for the development of two-line system hybrid rice. Later, Yang & Wang [2] induced a mutant (5460 S) in the *indica* variety IR 54 that exhibited either pollen sterility or pollen fertility depending on change of temperature. This is called temperature sensitive genic male sterility. These two pioneer findings laid a new strategy of hybrid rice breeding, which involves in

exploitation of environmental genic male sterility. TGMS lines will be sterile when the temperature could be high (32 °C/24 °C), the same line become revert to fertile when the temperature will be low (24 °C/18 °C) [3]. Hence seed multiplication of TGMS lines could be possible the place where low temperature prevails. In the tropical countries like India where temperature fluctuations are common, the thermosensitive genic male sterility (TGMS) system can be effectively utilized. Temperature sensitive genic male sterility (TGMS) and wide compatible varieties provide a new tool for direct utilization of rice intersubspecific heterosis by two line system [4]. Considering the potential of these novel approaches in hybrid development programme, the present investigation was attempted to develop two line rice hybrids involving TGMS lines in rice.

¹ Assistant Professor (PBG), Dryland Agricultural Research Station, Chettinad, Karaikudi, Sivaganga District-630 102, Tamil Nadu, India, *E-mail: sasikalacpbg@gmail.com

² Assistant Professor, UG Botany lab, Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore-641 003, Tamil Nadu, India

³ The Dean, Forest College and Research Institute, Mettupalayam, Tamil Nadu, India

MATERIALS AND METHODS

Present investigation was carried out in Paddy Breeding Station, Centre for Plant Breeding and Genetics, TamilNadu Agricultural University, Coimbatore, India. Experimental material which consists of four TGMS lines *viz.*, TNAU 27S, TS09 12, TS09 15, TS09 25 and 15 non TGMS pollen parents (Table 1) and sixty hybrids. TGMS lines and testers were raised for crossing programme during summer 2010 (April-May) when maximum/minimum temperature was prevailed 34.52-36.55 °C/25.05-25.19°C. Crossing was done by adopting clipping method. Field evaluation of 79 genotypes (19 parents and 60 hybrids) along with two checks CORH 3 and Improved white ponni in Randomized Block Design with two replications during *kharif* 2010. The performance of F₁ hybrid combinations were evaluated by recording observations on the following attributes as per the IRRI, Standard Evaluation System (1996)[5]. At flowering stage in each replication, five competitive plants in the middle rows were selected for taking following biometrical observation. There are totally nine quantitative characters were recorded *viz.*, days to fifty per cent flowering, plant height, number of productive tiller, panicle length, number of spikelets/panicle, number of filled spikelets/panicle, spikelet fertility percentage, 1000 grain weight and single plant yield.

Table 1
Lines and testers used for hybridization programme

Sl.No.	Lines	Sl.No.	Testers
1	TNAU 27 S	1	CO 47
2	TS 09 12	2	CO 50
3	TS 09 15	3	CB 05/501
4	TS 09 25	4	ADT 39
		5	ADT 43
	Checks	6	BPT 5204
1	CORH 3	7	KDML 105
2	Improved white ponni	8	WGL 14
		9	G 14
		10	RJ 101
		11	RJ 102
		12	RJ 103
		13	RJ 104
		14	CB 05 911/884
		15	CB 87 R

The mean data for each character individually was subjected to statistical analysis. The total variance was partitioned into various sources by following the model suggested by Panse & Sukhatme [6], combining ability analysis was carried out according to Kempthorne [7]. The percent increase or decrease of

F₁ hybrids over parents was assessed as heterotic effects by following Fonesca & Patterson [8].

RESULTS AND DISCUSSION

Analysis of variance for different quantitative traits

Analysis of variance (ANOVA) for combining ability studies revealed that significant difference in among lines, testers and hybrids. Variance due to lines x tester was significant for all quantitative traits. The SCA variance was higher than the GCA variance in all yield contributing characters except total productive tillers (Table 2). This indicated that the presence of non additive gene action governing the traits. Preponderance of non additive gene action in yield and yield attributing traits indicated that need to exploitation of heterosis for further improvement. Many reports are available in rice for combining ability and gene action on various characters including yield [9, 10, and 11].

General combining ability effects and *per se* performance of parents

Various biometrical techniques have been successfully used by to assess the genetic makeup of different lines. The General combining ability effects (*gca*) for each quantitative trait was given in Tables 3.

The estimates of *gca* effects showed that parents with high *gca* effects differ for various traits. Among four TGMS lines, TNAU 27 S was judged as good general combiner for traits *viz.*, days to fifty per cent flowering (-0.958), number of productive tiller (3.021), panicle length (0.632), thousand grain weight (2.009) and single plant yield (3.455) (Tables 3). TS 09 25 was good combiner for increasing number of spikelets per panicle (21.736) and number of filled spikelets per panicle (17.114). Hence, these two TGMS lines were identified for generating of good hybrid combinations.

Among testers CO 47, CO05/501, KDML 105, BPT 5204, WGL 14 and RJ 102 registered positive *gca* effects for various yield and yield contributing traits. Among them KDML 105 had registered high *gca* for single plant yield (4.3) and positive *gca* effect for thousand grain weight. WGL 14 showed positive *gca* for number of spikelets per panicle and higher *gca* effect for number of filled spikelets (22.478) per panicle RJ 102 had positive alleles for spikelet fertility per cent (5.502) and thousand grain weight (3.813) by registering high *per se* and positive significant *gca* effect. Best general combiners with respect to grain yield per plant were reported by scientists [12, 13]. CB 05/501, KDML 105 and CO 47 genotypes expressed high *per se* and

Table 2
Analysis of variance for combining ability for different quantitative traits

Source of variance	df	Day to 50% flowering	Plant height (cm)	Total productive tillers	Panicle length	Number of spikelets/panicle	Number of filled spikelets/panicle	Spikelets fertility %	1000 grain weight	Single plant yield
Crosses	59	45.381**	397.674**	24.032**	7.533**	2153.443**	1566.527**	112.105**	22.904**	43.760**
Lines	3	67.631	356.364	248.992**	7.512	8973.293**	7744.974**	20.961	64.801**	288.942**
Testers	14	36.169	1060.477**	12.724	8.155	3168.892*	2183.194*	190.234*	39.984**	36.604
L x T	42	46.863**	179.691**	11.733**	7.327**	1327.827**	919.654**	92.573**	14.218**	28.632**
Error	59	0.669	3.089	2.716	1.39	294.541	133.895	45.258	0.676	1.181
Total	119	23.269	198.698	13.267	4.438	1217.379	846.73	78.404	11.691	22.286
σ^2 GCA		0.265	27.828**	6.269**	0.0266*	249.646**	212.865**	0.686*	2.009**	7.060**
σ^2 SCA		23.084**	87.834**	4.6195**	3.053**	542.705**	398.696**	24.364**	6.787**	13.622**
σ^2 GCA / σ^2 SCA		0.012	0.317	1.357	0.009	0.46	0.534	0.028	0.296	0.518

** Significant at 1% level

* Significant at 5% level

Table 3
General combining effects (gca) of lines and testers

S.No.	Parents	Days to 50% flowering	Plant Height (cm)	Total Productive Tillers	Panicle length (cm)	Number of Spikelets/panicle	Number of Filled Spikelets/panicle	Spikelet Fertility %	1000 grain weight (g)	Single plant yield (g)
Lines										
1	TNAU 27 S	-0.958**	-1.141**	3.021**	0.632**	-20.030**	-20.226**	-0.032	2.009**	3.455**
2	TS 09 12	-1.525**	-0.37	1.898**	-0.575**	2.48	7.373**	-0.493	-1.324**	-1.197**
3	TS 09 15	0.775**	-3.310**	-2.648**	0.071	-4.186*	-4.261**	1.188	-0.820**	-3.676**
4	TS 09 25	1.708**	4.821**	-2.271**	-0.128	21.736**	17.114**	-0.663	0.135	1.418**
Testers										
1	CO 47	-1.117**	-9.001**	-0.127	-2.026**	7.634	8.729*	2.025	-2.407**	2.862**
2	CO 50	3.758**	-0.286	-0.46	0.57	-11.616*	-0.191	3.56	-0.640*	0.782
3	CB 05/501	-0.242	-7.635**	-0.043	-1.172**	17.175**	21.228**	1.574	-0.570*	2.497**
4	ADT 39	1.258**	-2.788**	0.858	0.717	17.674**	9.353*	-2.175	0.866**	0.157
5	ADT 43	-0.242	-4.371**	0.208	0.952*	8.301	0.311	-1.684	-3.119**	-2.418**
6	BPT 5204	1.008**	-2.785**	0.673	2.324**	2.967	13.897**	3.625	-3.363**	-0.229
7	KDML 105	0.008	12.477**	-1.043	0.103	5.592	6.563	-1.551	1.656**	4.300**
8	WGL 14	2.383**	-3.791**	1.624**	-0.313	23.759**	22.478**	2.436	-2.708**	-0.2
9	G14	0.758*	3.395**	-1.098	-0.329	11.426*	5.853	-0.358	1.900**	0.631
10	RJ 101	0.883**	-5.430**	-0.293	0.268	-21.533**	-31.781**	-11.515**	2.578**	-1.063*
11	RJ 102	-4.867**	31.510**	-0.627	-0.355	-12.910*	-6.021	5.502*	3.813**	0.327
12	RJ 103	-1.367**	-8.155**	-2.514**	0.207	-42.033**	-30.022**	2.144	-0.753*	-3.173**
13	RJ 104	-2.617**	15.412**	1.527**	0.385	-21.409**	-4.771	7.135**	0.513	-1.023*
14	CB 05 911/884	1.883**	-8.350**	2.372**	-0.944*	29.549**	5.187	-7.441**	-0.198	-0.01
15	CB 87 R	-1.492**	-10.205**	-1.056	-0.387	-14.576*	-20.813**	-3.276	2.432**	-3.438**
	S.E. (Lines)	0.1521	0.3662	0.2883	0.2019	2.8426	2.0188	1.2089	0.1466	0.2151
	S.E. (Testers)	0.2946	0.7091	0.5584	0.3909	5.5047	3.9093	2.341	0.2839	0.4165
	S.E. (Hybrids)	0.5892	1.4183	1.1167	0.7818	11.0095	7.8186	4.6821	0.5678	0.833

** Significant at 1% level * Significant at 5% level

positive significant *gca* for single plant yield. Hence, TNAU 27 S, TS09 25, KDML 105, CB 05/501 and CO 47 are good general combiners for yield and yield related traits and these lines may possess with favorable alleles for traits which mentioned above and these lines can be effectively utilized for hybridization programme as potential donors for exploitation of good commercial rice hybrids.

Specific combining ability (*sca*) effects of hybrids

Top ranking five hybrids with standard heterotic values for two checks, mean performance, *sca* effect and *gca* effect of parents were represented as high, medium and low (Table 4).

In the present investigation, top ranking hybrids on each character showed any one of the parent pertaining with high *gca* effect except the hybrid TS 09 15 x CO 50 for panicle length (Table 4) this indicated that involvement of both poor combiners also produced superior specific combining hybrids. Involvement of both combiners with low *gca* has been attributed to over dominance or epistatis interaction which has been suggested by Dalvi & Patel [14]. For thousand grain weight, the hybrids TNAU 27 S x RJ 103 and TNAU 27 S x CB 05 911/884 registered highly significant specific combining ability effect and good mean performance with high x medium and high x low general combining ability effect respectively. Cross combinations involving parents with the combining ability of High x High is due to positive alleles of both parents. And also reports are showed that cross combinations involving high x high combiners which can be fixed in subsequent generations if there is absent repulsion phase of linkages [15].

For grain yield crosses TNAU 27S x CB05 911/884 and TNAU 27S x BPT 5204 recorded high x low *gca* effects parents with high *sca* effects were result of interaction between positive allele from good combiner and negative allele from poor combiner. This result has inferred that high *sca* effects of crosses comprising additive and dominance components of gene action where as, high *sca* effects of crosses involving low x low (TS 09 15 x CO 50) combiners might be attributed to dominance x dominance type of gene action [11, 13]. TS09 25 x BPT 5204 showed positive and high *sca* effect for panicle length (2.346), number of spikelets per panicle (39.722) and number of spikelets per panicle (36.217) with low x high, high x low and high x high *gca* interactions of parents respectively. This hybrid provide a the clear-cut picture of gene interaction of same cross combination

in different yield contributing characters can be useful to breeder for exploiting of superior heterotic hybrids.

TNAU 27 S x KDML 105 had noticed for high specific combining ability effects for seven traits *viz.*, days to fifty per cent flowering (-3.542), plant height (25.026), total productive tillers (3.229), panicle length (2.173), number of spikelets per panicle (24.198), 1000 grain weight (1.245) and single plant yield (6.636).

Heterosis and *per se* performance of hybrids

Heterosis and *per se* performance for five top ranking hybrids for each was presented in each trait (Table 4). Negative standard heterosis over the check improved white ponni was observed for days to fifty percent flowering in hybrids *viz.*, TS09 12 x RJ 104, TS09 12 x KDML 105, TS09 12 x BPT 5204, TS09 12 x RJ 103 and TNAU 27S x RJ 102. Result indicated that improvement of early cultivars through heterosis breeding. Three hybrids such as TS 09 25 x BPT 5204, TS 09 25 x ADT 43 and TS 09 25 x ADT 39 were observed for high mean performance and high heterosis over two checks for two traits *viz.*, number of spikelets per panicle and number of filled spikelets per panicle.

Standard heterosis (>20%) over CORH3 and more than 55 per cent over improved white ponni was observed in hybrids TS09 25 x RJ 101 and TNAU 27 S x G 14 for thousand grain weight. The prime objective of hybrid development is obtaining superior hybrids coupled with high yield and the present study highlighted the best performing hybrids for yield improvement *viz.*, TNAU 27 S x KDML 105, TS 09 25 x KDML 105, TNAU 27 S x CB 05 911/884, TNAU 27 S x BPT 5204 and TNAU 27 S x CB05/ 501 with good *per se* performance, high specific combining ability, good general combining ability of parental lines (High x High and High x Low) and highest per cent of standard heterosis over two checks (CORH 3 and Improved white ponni). According to Peng & Virmani [16] the possibility of interaction between positive alleles from good combiner and negative alleles from poor combiner in high x low cross combination and suggested for the exploitation of F₁ generation, as their high yielding potential would be unfixable in succeeding generation.

Among five hybrids selected for single plant yield, TNAU 27 S x KDML 105 registered the all positive aspects a typical hybrid with which early mid duration, spikelet fertility of 76.55 per cent and high per cent of standard heterosis for single plant yield as 51.94 per cent over CORH 3 and 87.71 per cent over improved white ponni. Based on suggestions many

Table 4
Selected hybrids with high *Per se*, standard heterosis, *sca* and *gca* effect of parents

<i>Characters/Hybrids</i>	<i>Per se</i>	<i>diii (1) over CORH 3</i>	<i>diii (2) over IWP</i>	<i>sca</i>	<i>gca effect of parents</i>
Days to 50% flowering					
TS 09 12 x RJ 104	82.50	1.23	-28.88**	-3.35**	High x High
TS 09 12 x KDML 105	82.50	1.23	-28.88**	-5.975**	High x Low
TS 09 12 x BPT 5204	83.00	1.84	-28.45**	-6.475**	High x Medium
TS 09 12 x RJ 103	83.50	2.45*	-28.02**	-3.60**	High x High
TNAU 27 S x RJ 102	83.58	2.45*	-28.02**	-0.667	High x High
Plant height					
TS 09 15 x CB 05 911/884	79.25	-3.65	-25.73**	-7.899**	High x High
TNAU 27 S x ADT 43	81.92	-0.41	-23.23**	-11.382**	High x High
TNAU 27 S x RJ 103	82.92	0.81	-22.29**	-6.598**	High x High
TNAU 27 S x BPT 5204	83.30	64.34**	26.68**	-11.553**	High x High
TS 09 12 x RJ 103	83.84	1.93	-21.43**	-6.448**	Low x High
Total productive tillers					
TNAU 27 S x BPT 5204	23.45	65.53**	174.27**	4.963**	High x Low
TNAU 27 S x CB 05 911/884	23.33	64.68**	172.87**	3.144**	High x High
TS 09 12 x CB 05 911/884	21.67	52.93**	153.39**	2.602*	High x High
TS 09 12 x CO 47	20.50	44.70**	139.77**	3.935**	High x Low
TNAU 27 S x WGL 14	20.17	42.37**	135.91**	0.732	High x High
Panicle length					
TS 09 25 x BPT 5204	27.99	41.70**	26.09**	2.346**	Low x High
TS 09 15 x CO 50	27.07	37.04**	21.94**	2.982**	Low x Low
TS 09 15 x ADT 43	26.65	34.91**	20.05**	2.179**	Low x High
TNAU 27 S x WGL 14	26.38	33.57**	18.86**	2.618**	High x Low
TNAU 27 S x KDML 105	26.35	33.42**	18.72**	2.172**	High x Low
Number of spikelets / panicle					
TS 09 12 x CB 05 911/884	238.17	54.99**	17.66*	34.061**	Low x High
TS 09 25 x BPT 5204	236.50	53.90**	16.84	39.722**	High x Low
TS 09 25 x ADT 43	223.80	52.93**	16.10	32.888**	High x Low
TS 09 15 x G 14	226.00	47.07**	11.65	46.685**	Medium x High
TS 09 25 x ADT 39	223.83	45.66**	10.58	12.345	High x High
Number of filled spikelets/ panicle					
TS 09 25 x BPT 5204	219.00	68.68**	20.99**	36.217**	High x High
TS 09 25 x ADT 39	206.66	59.18**	14.18*	28.426**	High x High
TS 09 25 x CO 47	199.67	53.79**	10.31	22.055**	High x High
TS 09 25 x ADT 43	196.50	51.35**	8.56	27.303**	High x Low
TS 09 25 x KDML 105	189.34	45.83**	4.60	13.886	High x Low
Spikelet fertility per cent					
TS 09 25 x RJ 104	99.87	18.19*	11.67	6.633	Low x High
1000 grain weight					
TS 09 25 x RJ 101	27.01	23.68**	59.60**	3.836**	Low x High
TNAU 27 S x G 14	26.39	20.84**	55.94**	2.021**	High x High
TS 09 25 x RJ 102	26.20	19.99**	54.85**	1.796**	Low x High
TNAU 27 S x RJ 103	25.84	18.37**	52.75**	4.134**	High x Medium
TNAU 27 S x CB 05 911/884	25.80	18.16**	52.48**	3.534**	High x Low
Single plant yield					
TNAU 27 S x KDML 105	44.00	51.94**	87.71**	6.636**	High x High
TS 09 25 x KDML 105	40.92	41.29**	74.55**	5.589**	High x High
TNAU 27 S x CB 05 911/884	38.80	33.99**	65.53**	5.746**	High x Low
TNAU 27 S x BPT 5204	37.00	27.77**	57.85**	4.165**	High x Low
TNAU 27 S x CB05/ 501	35.81	23.66**	52.77**	0.249	High x High

scientists, hybrids with 20-30% positive standard heterosis over commercial varieties is sufficient to offset the additional cost of hybrid seeds for self pollinated crops [17, 18].

Effects of *gca* combined with *per se* performance of each line revealed that the actual potentiality of lines. The study revealed that the TNAU 27S, TS 09 25, KDML 105 and CB 05/501 were not only good general combiners for single plant yield but also produced high heterotic combination of hybrids *viz.*, TNAU 27S x KDML 105 (44g), TS 09 25 x KDML 105 (40.92) and TNAU 27S x CB 05/501 (38.80) with high mean performance for single plant yield. A potential hybrid is one that has superior performance, which is the actual realized value. The realized mean value should be above the standard parent's performance for commercial exploitation. Hence, it would be fruitful if the hybrids are evaluated based on the mean values, standard heterosis and *sca* effect [13, 15, 19]. Among hybrids, TNAU 27S x KDML 105, TS 09 25 x KDML 105, TNAU 27 S x CB 05 911/884, TNAU 27 S x BPT 5204 and TNAU 27 S x CB 05/ 501 was recommended for its high per cent of standard heterosis for single plant yield and high specific combining ability effects and good *per se* performance. Hence, exploitation of these hybrids for commercialization will lead to change the current hybrid era.

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