Oryza Vol. 54 No. 4, 2017 (401-406)
DOI 10.5958/2249-5266.2017.00054.6

Critical stages of thermo-sensitive genic male sterile lines of rice in Kerala

Rajesh T[1]*, Radhakrishnan VV[4], Pressana Kumari KT[1], Rose Mary Francies[1], Sreenivasan E[2], Ibrahim KK[1] and Latha A[3]

[1]*College of Horticulture, Vellanikkara, Kerala Agricultural University, Thrissur, Kerala, India*

[2]*Agronomic Research Station, Chalakudy, Kerala Agricultural University, Thrissur, Kerala, India*

[3]*Agricultural Research Station, Mannuthy, Kerala Agricultural University, Thrissur, Kerala, India*

[4]*Kerala Agricultural University, Thrissur, Kerala, India*

**Corresponding author e-mail: atr.agri@gmail.com*

Received : 10 February 2017 Accepted : 1 December 2017 Published : 20 December 2017

ABSTRACT

Two line breeding is a viable proposition in a state like Kerala where rice is cultivated from below mean sea level to altitudes of 1500 MSL. The system provides a much simpler and economic hybrid seed production and broader choice of male parents for enhancing yield potential. A set of seven promising Temperature Sensitive Genic Male Sterile lines received from Tamil Nadu Agricultural University, Coimbatore, India viz., TGMS 74S, TGMS 81S, TGMS 82S, TGMS 91S, TGMS 92S, TGMS 93S and TGMS 94S were characterized for their sterility, fertility behavior under field conditions. Based on sterility /fertility behavior the entire study period of 2011 March to 2012 March can be grouped as two distinct periods viz, pollen sterile and pollen fertile months. The pollen sterile months indicated April and May when 100 per cent sterility was observed. July and August can be considered as pollen fertile months. Other months exhibited un stable expression of sterility /fertility and hence could not be grouped under pollen sterile or pollen fertile months. The sterile period was longest in TGMS 82S (87 days) followed by TGMS 81S (83 days). Since all these lines showed completely sterile for more than 30 consecutive days during sterile phase, these lines were successfully utilized in hybrid rice breeding programme. The Critical Sterility Temperature ranged from 34.2^{0}C (TGMS 74S, TGMS 91S, and TGMS 94S) to 38.6^{0}C (TGMS92S). Critical fertility temperature ranged from 23.6 ^{0}C (TGMS 91S) to 25.4^{0}C (TGMS 92S, TGMS 93S).

Key words: *Oryza sativa, TGMS, fertility behavior, CST, CFT*

INTRODUCTION

Kerala, 'God's own country' occupies a unique position on the map of the country and lies between 8^{0}18' and 12^{0}48' North latitude and between 74^0 52' and 77^0 24' East longitude. It has a total area of 38,855 km^2. Rice is the staple food of the people of Kerala, and, traditionally, the cultivation of rice has occupied pride of place in the agrarian economy of the State. The lush green of paddy fields is one of the most captivating features of Kerala's landscape. The state's different regions grow paddy in different ways. While the crop is cultivated two to three metres above the sea level in Kuttanad, it is grown above 1,400 metres in Wayanad district on the Western Ghats. In the coastal regions of central Kerala, Pokkali, a special salt-tolerant rice variety, is alternated with prawn in saline water. The state also has some medicinal and highly nutritional rice varieties.

For the last several decades in Kerala the area under rice cultivation and rice production has shown more or less a consistent fall and rice cultivation faces stiff competition from crops like banana, ginger, rubber, coconut and mango. With increasing population, demand for increased production of rice must be met from less land with limited resources. Hybrid rice technology is the best option for increasing the production of rice in tropics (Virmani, 1996). For heterosis exploitation in rice, the two-line hybrid rice system shows many advantages over the three-line system of hybrid rice, including greater ease in determining the pollinator line in the TGMS male sterility system and a 5 to 15 percent higher heterosis.

The foremost step to start two line breeding is characterization of TGMS lines and determination of fertility behavior of particular line to find out critical sterility temperature (CST) and critical fertility temperature (CFT). Because TGMS lines are sterile in certain temperature regime there is need to determine CST and CFT. Hence the determination of CST and CFT will provide a broad way to decide suitable season and locations for TGMS seed multiplication and hybrid seed production (Latha et al., 2004). TGMS lines greatly differ in their sensitivity to temperature fluctuation. Only those lines with lower critical sterility point and those, which do not revert back to fertility with slight decrease in temperature, are useful to develop two line hybrids (Wu, 1997). This critical sterility temperature (CST) and critical fertility temperature (CFT) were found to vary in different TGMS lines as the TGMS gene of these lines are transferred into different genetic background.

The lowest mean temperature at which the line becomes sterile from the fertile condition is the critical sterility temperature (CST). The highest mean temperature at which the line becomes fertile from the sterile condition is the critical fertility temperature (CFT). The critical temperature inducing sterility is relatively low (23º C) in temperate zone and 24º C in sub-tropics (Yuan, 1998). Hence, the present study was undertaken with the specific objective of characterizing a set of promising TGMS lines for their critical stages so as to use them in two-line heterosis breeding.

MATERIALS AND METHODS

A set of 7 TGMS lines received from Tamil Nadu Agricultural University (TNAU), Coimbatore, India (Table 2) were evaluated for pollen and spikelet fertility during March 2011 to 2012 March at COH Vellanikkara (High temperature condition - 36/22°C) and CRS Pampadumpara (Low temperature condition - 30/13°C) simultaneously to identify the critical stages of TGMS lines.

Table 1. IRRI Standard Evaluation System (1996).

Pollen sterility (%)	Group
100.00	Completely sterile
99.00 - 99.90	Highly sterile
95.00 - 98.90	Sterile
70.00 - 94.90	Partially Sterile
< 70	Partially fertile to Fertile

The TGMS lines were raised at fortnightly interval from March, 2011 to March, 2012. Standard package of practices were carried out throughout the crop period as recommended in the Package of Practices Recommendations: Crops, Kerala Agricultural University (2010). Pollen sterility/fertility behavior were studied in relation to weather parameters collected from the department of meteorology, COH Vellanikkara and CRS Pampadumpara (Table 3). The pollen sterility/fertility was observed in three randomly selected plants at the time of heading. The spikelets were collected before anthesis, the anthers were dissected out from the spikelet, squeezed to release the pollen grain and stained using I-KI (1%) solution. Darkly stained and round pollen grains were considered as fertile, whereas irregular shaped non stained pollen grains were grouped as sterile pollen. Pollen fertility/sterility per centage was worked out.

$$\text{Pollen fertility (\%)} = \frac{\text{Number of stained round pollen}}{\text{Total number of pollen counted}} \text{x}100$$

The sterility status was fixed as under following the IRRI Standard Evaluation System (1996) (Table 1). From the one year data on pollen sterility/fertility the fertile and sterile phases of TGMS lines, the duration of each phase and the fertility transition phase were identified. The flowering period in which the line with full sterility (100 per cent pollen sterility) were taken at sterile phase. The period in which the plants recorded more than 50 per cent pollen fertility was considered as fertile phase. The period of partial sterility was

Table 2. TGMS lines taken for evaluation.

Sl.No	TGMS line	Source
1	TGMS 74S	TNAU , Coimbatore
2	TGMS 81S	TNAU, Coimbatore
3	TGMS 82S	TNAU, Coimbatore
4	TGMS 91S	TNAU, Coimbatore
5	TGMS 92S	TNAU, Coimbatore
6	TGMS 93S	TNAU, Coimbatore
7	TGMS 94S	TNAU, Coimbatore

considered as the phase of fertility transition.

RESULTS AND DISCUSSION

Among the seven TGMS lines,the TGMS lines TGMS 74S, TGMS 81S, TGMS 82S and TGMS 94S showed completely male sterile (100 per cent) during April first week to June second week. TGMS 91S, TGMS 92S, TGMS 93S showed completely male sterile (100 per cent) from April second week to June second week. On visual inspections white coloured non dehiscent anthers were noted and no seed set was observed on bagging the panicles. June third week onwards slowly change to fertile condition to partial fertile and all TGMS lines showed 100% fertile from July second week to August third week. During this period the anthers were yellow and dehiscent. Seeds were produced in the bagged panicles. Fertile pollen grains which were spherical and stained were noted mixed with lightly stained and spherical unstained pollen grains.

Sterility/fertility behaviour

The two-line system simplifies the production of hybrid, since only pair of pure fertile and sterile lines are required. It can also eliminate the potential negative effects associated with the CMS. Furthermore, the nuclear genes responsible for sterility are relatively easy to be transferred to diverse genetic background. However, owing to the limitation of the temperature and/or photoperiod requirement, an EGMS line can only be used in a relatively narrow zone, and suitable sterile lines must be developed for a target production environment (He et al., 2006). Therefore characterization of TGMS lines with respect to their fertility / sterility alteration behaviour will provide clear cut idea of utilization of that particular line for predicting appropriate timings for hybridization programme (at sterile phase) as well as seed multiplication (at fertile phase).

Based on sterility /fertility behavior, the entire study period of 2011 March to 2012 March can be grouped as two distinct periods *viz.,* pollen sterile and pollen fertile. In the present study, the pollen sterile months indicated April, May when 100 per cent sterility was observed. July and August can be considered as pollen fertile months. Other months exhibited un stable expression of sterility/fertility and hence could not be grouped under pollen sterile or pollen fertile months. Sterility/fertility behaviour of pollen grains over months during the study period in two locations was sown in Fig. 1 and Fig. 2.

The sterile period was longest in TGMS 82S (87 days) followed by TGMS 81S (83 days). Since all these lines were completely sterile for more than 30 consecutive days during sterile phase, hybrid seed production utilizing these lines can be taken up at COH Vellanikkara by raising the lines in such a way that following coincides with the sterile phase. All the lines reverted in to fertile from July 2nd week to September 1st week., All lines exhibited 100 per cent pollen fertility during the periods. Since all these were completely fertile for more than 30 consecutive days during fertile phase multiplication of TGMS lines with purity standard will be easier for these lines. Lu et al. (1998) suggested that for successful utilization of TGMS lines the sterile

Table 3. Monthly weather data of COH Vellanikkara and CRS Pampadumpara (2011-2012).

Month	Maximum Temperature (^{0}C)		Minimum Temperature (^{0}C)		Relative humidity (%)		Sunshine hours		Rainfall (mm)	
	COH Vellanikkara	CRS Pampadumpara	COH Vellanikkara	CRS Pampadumpara	COH Vellanikkara	CRS Pampadumpara	COH Vellanikkara	CRS Pampadumpara	COH Vellanikkara	CRS Pampadumpara
April-2011	34.3	27.7	24.5	18.6	58	77.0	6.6	6.0	207.1	178.8
May	33.0	27.6	24.9	19.4	63	78.0	6.8	7.5	198.5	24.0
June	29.3	23.0	23.6	18.6	82	91.9	2.5	2.5	799.6	360.4
July	29.1	22.3	22.5	18.1	81	93.5	1.6	1.2	500.2	10.9
August	29.4	23.0	22.9	18.0	78	92.6	2.2	1.5	713.0	11.3
September	30.0	24.0	23.1	17.8	75	87.4	5.4	3.9	435.2	151.4
October	31.1	26.3	23.5	18.9	65	95.0	6.1	4.8	193.0	12.6
November	31.4	23.9	22.9	17.3	57	83.8	6.3	5.1	240.0	184.0
December	31.9	24.1	21.9	16.8	49	79.6	7.3	5.6	002.4	21.0
January-2012	32.8	24.1	21.3	15.1	58	71.7	9.5	8.6	000.0	6.8
February	35.1	26.3	22.1	16.0	54	64.4	9.2	8.4	000.0	0.0
March	35.2	29.1	24.2	18.6	67	66.4	7.6	7.2	003.5	2.4

Table 4. Correlation coefficient of pollen sterility with mean of different weather parameters over the period of 26 days before heading.

Weather parameters	TGMS 74S	TGMS 81S	TGMS 82S	TGMS 91S	TGMS 92S	TGMS 93S	TGMS 94S
Maximum temperature	0.507**	0.415*	0.397*	0.338*	0.305	0.540**	0.585**
Minimum temperature	0.306	0.430*	0.584**	0.392*	0.423*	0.335*	0.397*
Mean temperature	0.566**	0.391*	0.415*	0.385*	0.368*	0.586**	0.675**
Relative humidity	-0.092	-0.063	0.078	-0.097	0.123	-0.086	-0.109
Rainfall	-0.002	0.215	0.237	-0.057	-0.098	0.015	0.161
Sunshine hours	0.294	-0.273	-0.273	0.286	0.418*	0.168	0.365*

Table 5. Critical temperature for TGMS lines.

Entries	Heading date with sterility	Duration	Pollen sterility(%)	CST (^{0}C)	Heading date with maximum fertility	Duration	Pollen fertility (%)	CFT (^{0}C)
TGMS 74S	Apr 01-June 18	79	100	34.2	July 06 -Aug31	57	100	23.0
TGMS 81S	Mar 25-June 14	83	100	32.9	July 10-Aug 31	53	100	24.2
TGMS 82S	Mar 25 -June 18	87	100	32.9	June 28-Aug 28	52	100	24.2
TGMS 91S	Apr 05-June 15	72	100	34.2	July 10- Aug 30	52	100	22.7
TGMS 92S	Apr 08-June 15	69	100	34.2	July 10- Aug 31	53	100	24.2
TGMS 93S	Apr 01-June 17	78	100	34.2	July 10- Sep 01	54	100	24.2
TGMS 94S	Apr 01 -June 18	79	100	34.2	July 11 -Sep 01	53	100	24.2

and fertile phase should be atleast for 30 consecutive days. Similar findings was reported by latha et al. (2004) in TS6 completely sterile for 78 consecutive days and reverted to fertile for continuously 69 days in the same Coimbatore location.

Influence of weather factors on fertility alteration

Relative influence of primary weather factors such as maximum and mean temperature secondary weather factors such as relative humidity, rainfall and sunshine hours on pollen sterility/fertility alteration behavior will vary among different lines due to different source of male sterility genes and genetic backgrounds (Wu et al., 1991; Zhang and Lu, 1991). Present investigation revealed that the influence of maximum and mean temperature on pollen sterility for all TGMS lines.

Correlation studies between pollen sterility and different weather factors showed that the influence of primary as well secondary weather factors act on fertility alteration (Table 4).

In the present study, all seven TGMS lines

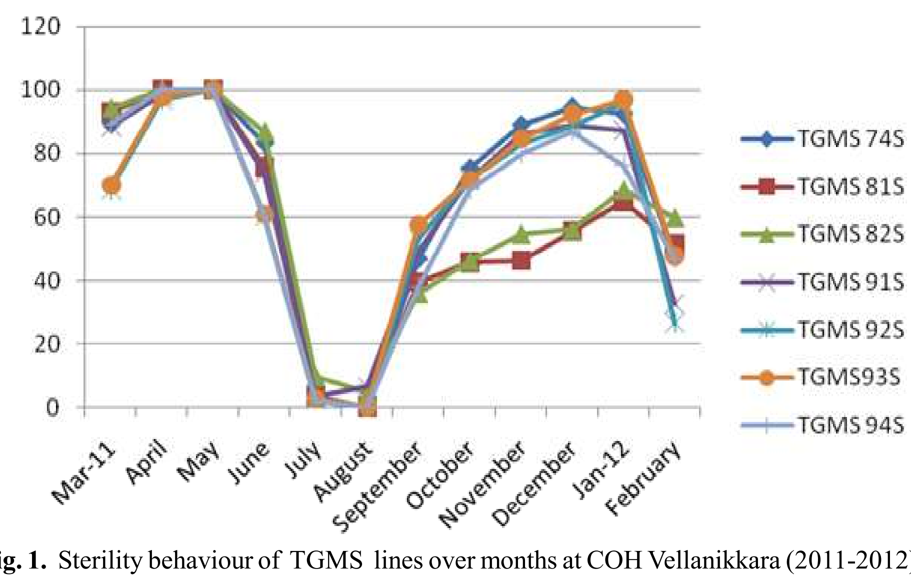

Fig. 1. Sterility behaviour of TGMS lines over months at COH Vellanikkara (2011-2012).

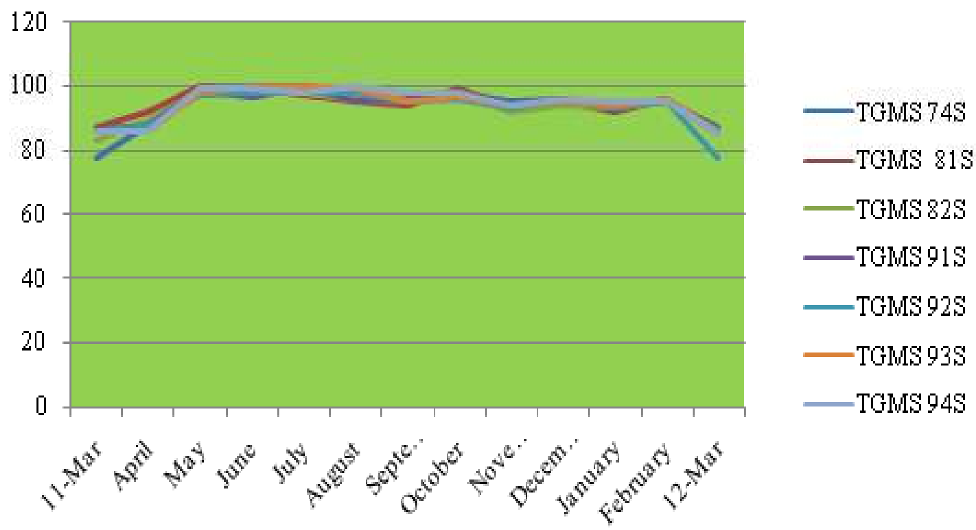

Fig. 2. Fertility behavior of TGMS lines over month at CRS Pampadumpara (2011-2012)

exhibited relative influence of maximum and mean temperature on pollen sterility with positively significant values. The significant influence of maximum temperature on pollen sterility was reported by Zhang and Lu (1991) and Chandirakala (2005). Except TGMS 74S, other six lines had significant influence of minimum temperature on pollen sterility. This is in accordance with the findings of Sun et al. (1989) and Liu et al (1997). In the present study, all the seven TGMS lines had no influence by secondary factors such as relative humidity and rainfall, on fertility alteration. This is in accordance with Zhang and Lu (1991) and Liu et al. (1997). Except TGMS 92S and TGMS 94S, all other TGMS lines had no influence on sunshine hours.

CST and CFT in TGMS lines

In the present investigation, under evaluation of seven TGMS lines the CST ranged from 34.2^{0}C (TGMS 74S , TGMS 91S and TGMS 94S) to 38.6^{0}C (TGMS92S). CFT ranged from 23.6^{0}C (TGMS 91S) to 25.4^{0}C (TGMS 92S, TGMS 93S) (Table 5). Similar findings also reported by Salgotra et al. (2012).

CONCLUSION

The results indicate that April and May seasons can be considered as the ideal period of exploiting 100 per cent sterility in rice for the hybrid seed production. Experiments at CRS Pampadumpara was the suitable location for Temperature Sensitive Genic Male Sterile line seed multiplication. The study concluded that CST(Critical Sterility Temperature) ranged from 34.2 to 38.6^0 C and CFT(Critical Fertility Temperature) ranged from 23.6 to 25.4^{0}C. Correlation studies between weather parameters and pollen sterility behavior indicated that maximum temperature and mean temperature had the highest significant positive correlation with sterility. All lines were not influenced by environmental factors such as relative humidity and rainfall.

REFERENCES

Ali J, Siddidq EA, Zaman FU, Abraham MJ and Ahemed I (1995). Identification and characterization of temperature sensitive genic amle sterile sources in rice (*Oryza sativa* L.). Indian J. Genet. 55: 243-259

Chandirakala R (2005). Genetic analysis and characterization of thermo sensitive genic male sterile lines (TGMS) and their utilization in heterosis breeding of rice (*Oryza sativa* L.), Ph.D. Thesis. Tamil Nadu Agricultural university, Coimbatore

He H, Peng X, Gong H, Zhu C and Ye G (2006). Fertility behaviour of rice (*Oryza sativa* L.) lines with dominant male sterile gene and inheritance of sterility and fertility restoration. Field crops Res. 98(1): 30-38

Lu XG, Virmani SS and Rencui Y (1998). Advances in Two line hybrid rice breeding.pp. 89-98. In: S. S.Virmani, E. A. Siddiq and K.Muralidharan (eds.). Advances in Hybrid Rice Technology. International Rice Research Institute, Manila, Philippines

Latha R, Thiyagarajan K and Senthilvel S (2004). Genetics, fertility behaviour and molecular marker analysis of new TGMS line TS 6 in rice. Plant Breed. 123: 235-240

Liu AM, Deng HF and Li BH (1997). Studies on fertility of Annong 810S. In: Proceeding of International Symposium on Two-line System of Heterosis Breeding in Crops.Sep.6-8, 1997, China National Hybrid Rice Research and development Centre, Changsha, China pp.188-192

Salgotra RK, Gupta BB and Ahmed MI (2012). Characterization of thermosensitive genic male sterility (TGMS) rice genotypes (*Oryza sativa* L.) at different altitudes. Aust J. of Crop Sci. 6(6): 957-962

Sun ZX, Xiong ZM, Min SK and Si SM (1989). Identification of temperature sensitive male sterile rice. Chinese J. Rice Sci. 3: 49-55

Virmani SS (1996). Hybrid rice. Adv. Agron. 57: 377-462

Virmani SS, Chaudary RC and Khush GS (1981). Current outlook on hybrid rice. Oryza 18 : 67-84

Viraktamath BC and Virmani SS (2001). Expression of thermosensitive genic male sterility in rice under varying temperature situations. Euphytica 122: 137-143

Wu XJ, Yin HQ and Yin H (1991). Preliminary study of temperature effect on Annong S1 and W6154S. Crop Res. 5: 4-6

Yuan LP (1998). Hybrid rice breeding in china. In: S.S.Viramani, E.A. Siddiq, and K. Muralidharan (Eds). Advances in hybrid rice technology. Proc. of the 3rd International Symposium on Hybrid Rice. 14-16 Nov. 1996 (Hyderabad, India). Int. Rice Res. Institute, Manila, Philippines pp. 27-33

Zhang XG and Lu XG (1991). Fertility response to photoperiod and temperature of five indica environment sensitive genic male sterile lines of rice. In: Agricultural Biotechnology. Yuu, C.B and Z.L.Chen (Eds). China Science and Technology, Press, Beijing, China pp. 678-679