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Studies on sterility behaviour in thermo-sensitive genic male sterile lines of rice

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Abstract
In hybrid rice, Thermo sensitive Genic Male Sterility (TGMS) system has several advantages compared to CMS system as it does not require maintainers for seed multiplication. In TGMS, the sterility/ fertility behavior is controlled by temperature. For successful utilization of TGMS lines in hybrid seed production, information on flowering behavior of the lines, critical stage of thermo sensitivity, critical sterility temperature, period of sterility and genotypic responses is indispensable. In this study, 25 TGMS lines from Paddy Breeding Station, Coimbatore were sown at weekly intervals from January 2, 2018 to January 31, 2018. Heading date (days), pollen sterility (%) and spikelet sterility (%) was recorded in each line for all the five staggered sowings. Information on maximum, minimum temperature and rainfall during the crop growth period was collected from meteorological observatory at the farm.
Based on flowering, 14 and 11 lines were identified to be medium and long in duration respectively. The sensitive stage for the TGMS lines was taken as 5 to 24 days before heading that coincided with the differentiation of secondary branch primordium and the filling stage of pollen. For the medium duration genotypes, critical stage fell between 65 and 99 days and for the long duration lines it fell between 80 and 112 days. The staggered sowings spread over a month had differential expression on days to heading among the TGMS lines which was narrow (< four days) in TNAU 14S, 18S, 71S and 116S and wider (12-10 days) in TNAU 136S, 139S and 137S. Altogether, 17 lines (eight medium and nine long duration) were 100% sterile in all the five staggerings that extended for a month. The critical sterility point (CSP) of the lines ranged from 28°C (TNAU 147S and TNAU 151S) to 31.7°C (TNAU 101S). Two other lines with low CSP were TNAU 45S and TNAU 71S. A total of 145mm rainfall received over a period of ten days during May 2018 which coincided with the sensitive stage in the last staggering of 10 TGMS lines resulted in the identification of 100% sterile lines with low CSP. Genotypic differences for expression of sterility existed even among the lines derived from same TGMS source.

Keywords: Hybrid rice, TGMS lines, flowering, Critical sterility point, Sensitive stage

Introduction
Commercial exploitation of heterosis has made tremendous achievement in hybrid rice breeding. Cytoplasmic Genic Male Sterility (CMS) system is used widely for hybrid seed production in rice. Lack or inadequacy of restorers is one of the major constraints in CMS and also male sterile cytoplasm has negative effect on yield heterosis (Shi *et al.,* 1996) [10]. The two – line system involving Thermo sensitive Genic Male Sterility (TGMS) in the tropics helps to overcome the constraints of CMS system as it does not require maintainers for seed multiplication. Any desirable line can be converted to TGMS line or can be used as a pollen parent, thus helps to widen the genetic base of hybrids (Viraktamath and Virmani 2001) [15]. Experiments and commercial practices have proved that two line breeding has 5-10% yield advantage over three - line breeding (Yuan, 1998) [18].
The sterility/ fertility behaviour of a TGMS line is controlled by temperature. The system is useful in tropical country like India where temperature variations between altitudes/seasons are available. But the major concern in two - line breeding is to ascertain that the TGMS line shows stable expression of sterility during hybrid seed production. The location and also time /season of sowing in a location is very crucial to achieve sterility over a prolonged period. In our earlier studies, Aduthurai and Coimbatore were identified as suitable locations for taking up two-line hybrid seed production (Sairekha *et al.,* 2017 and Vinodhini *et al.,* 2019) [7, 14]. In a particular location, determination of critical sterility point and duration of sterility of a TGMS line is important for its successful utilization.

In this context, the present study was undertaken at Paddy Breeding Station, Department of Rice to fix the critical phase and duration of sterility for a set of thermo sensitive genic male sterile lines.

Materials and Methods

Twenty five TGMS lines *viz.,* TNAU 14S, TNAU 18S, TNAU 45S, TNAU 46S, TNAU 67S, TNAU 71S, TNAU 83S, TNAU 84S, TNAU 86S, TNAU 93S, TNAU 100S, TNAU 101S, TNAU 114S, TNAU 115S, TNAU 116S, TNAU 124S, TNAU 135S, TNAU 136S, TNAU 137S, TNAU 139S, TNAU 142S, TNAU 143S, TNAU 145S, TNAU 147S and TNAU 151S developed/maintained at Paddy Breeding Station, Coimbatore were used as experimental material and their pedigree is given in Table 1.

Table 1: Pedigree details of twenty five TGMS lines in rice

S. No.	TGMS line	Pedigree	S. No.	TGMS line	Pedigree
1	TNAU 14S	IR 68301 S	14	TNAU 115S	TS-06-207-1-28
2	TNAU 18S	IR 75589-41-13-17-15-22 S	15	TNAU 116S	CBTS 282-6-22-1
3	TNAU 45S	GD 90489-3-29	16	TNAU 124S	TS 06-196-3-1
4	TNAU 46S	GD 99017-17	17	TNAU 135S	TS 29/ CO 49 (F_4 17S)
5	TNAU 67S	TS 29/ IR 62917-2-3-2R-1	18	TNAU 136S	TS 06-182-2
6	TNAU 71S	DRR 29S	19	TNAU 137S	TNAU 4S-1-2/CB 06-564
7	TNAU 83S	TNAU 45S-2	20	TNAU 139S	TS 29 100 Gy-3
8	TNAU 84S	TS 29 150Gy	21	TNAU 142S	TNAU 19S-1
9	TNAU 86S	MS-3	22	TNAU 143S	TNAU 19S-2
10	TNAU 93S	DRR 28S	23	TNAU 145S	TNAU 19S-4
11	TNAU 100S	ADT 39 100 Gy	24	TNAU 147S	TNAU 95S-1
12	TNAU 101S	DRR 23S-1	25	TNAU 151S	Not known
13	TNAU 114S	TNAU 4S-2/BPT5204			

Sowing of TGMS lines were taken up in weekly interval form January 2, 2018 to January 31, 2018 for five weeks. At 30 days, the lines were planted in single row, with 12 plants per row. The recommended package of practices was followed to ensure a healthy and good crop stand. In all staggerings, days to first flowering, pollen and spikelet sterility were recorded for the TGMS lines. For determination of pollen sterility, - anthers from five different plants in a line were squashed and pollen grains were stained with 1% Iodine Potassium Iodide (IKI) solution (Lopez and Virmani, 2000) [3]. Fertile pollen were round and darkly stained, while sterile pollen were irregular shaped, unstained or yellowish (Viraktamath and Virmani 2001) [15]. Three microscopic fields with total pollen count numbering 300 to 400 were observed and pollen sterility was expressed in percentage. For determining spikelet sterility, three plants were randomly selected for each genotype and spikelets were covered with glassine bags in order to prevent outcrossing. Spikelet sterility (%) was determined by counting the proportion of filled spikelets to the total number of spikelets (Lopez and Virmani, 2000) [3].
The following classification was adopted for categorization of lines based on sterility (SES, IRRI 2002) [12]

Table 2: The category of sterility percentage

Category	Sterility %
Completely Sterile	100
Highly sterile	99.0-99.9
Sterile	95.0-98.9
Partially Sterile	70.0-94.9
Fertile	< 70

The daily maximum and minimum temperature was recorded for 5 to 24 days before heading in all the lines in each staggering and the mean maximum and mean minimum temperatures for the critical period was arrived at for each of the lines. The temperature data was obtained from Paddy Breeding Station, Tamil Nadu Agricultural University (TNAU), Coimbatore.

Results and Discussion

Two line hybrid rice breeding is advantageous over three-line method which poses no restrictions for the restorer-maintainer relationship as in the case of CMS lines (Xinggui *et al.,* 1998) [17]. One of the constraints in adopting two- line hybrid rice breeding in a large scale is the presence of selfed seed in TGMS line during hybrid seed production due to temperature fluctuations. A sudden decrease in temperature during the sensitive period of panicle development causes the TGMS line to revert back to fertility, thus affecting the purity of seeds Thus we need to identify the temperature at which the line is completely sterile for its use in two line hybrid seed production (Sanchez and Viramani, 2005) [9].

Days to heading and sensitive stage for temperature

Critical Sterility Point (CSP) is the temperature at which particular line/genotype is completely sterile. The effect of temperature on male sterility/fertility is related to the developmental stage of the plant (Zongxiu *et al.,* 1993) [19]. Various researchers have reported the sensitive stage for temperature in their TGMS lines as follows. Zongxiu *et al.* (1993) [19] reported that formation of pollen mother cell to late uninucleate stage of pollen grains as the sensitive stage. Stamen pistil primordial stage was considered as sensitive stage by Ali *et al.* (1995) which is 15 -24 days before heading. Four to eight days after panicle initiation was considered as most sensitive stage for temperature variations by Viraktamath and Virmani (2001) [15]. Latha and Thiyagarajan (2010) reported that the sensitive stage for temperature was from stamen pistil primordial differentiation to pollen ripening. Differentiation of secondary branch primordium and the filling stage of pollen i.e., 24 to 5 days before heading was considered as sensitive stage for temperature by Sanchez and Virmani (2005) [9] while secondary branch primordial formation to stamen pistil primordial formation stage was considered as sensitive stage by Roy Stephen *et al*., (2016).
In our study, critical stage for sterility expression was considered as 5-24 days before heading that coincided with

the differentiation of secondary branch primordium and the filling stage of pollen. Accordingly, the sensitive stage fell between 65 and 112 days for the 19 lines which expressed sterility behavior in all or at least three staggerings (Table 3). For the ten genotypes which were medium in duration *viz.,* TNAU 46S, TNAU 67S, TNAU 100S, TNAU 115S, TNAU 116S, TNAU 124S, TNAU 137S, TNAU 139S, TNAU 142S and TNAU 143S, their critical stage fell between 65 and 99 days. For nine genotypes, *viz.,* TNAU 14S, TNAU 18S, TNAU 45S, TNAU 71S, TNAU 101S, TNAU 135S, TNAU 136S, TNAU 147S and TNAU 151S, the critical stage fell between 80 and 112 days.

Table 3: Critical stage for expression of pollen sterility in TGMS lines of rice

TGMS line	Critical stage (days)	TGMS line	Critical stage (days)
TNAU 46S	72-91	TNAU 14S	82-101
TNAU 67S	80-99	TNAU 18S	85-104
TNAU 100S	72-91	TNAU 45S	93-112
TNAU 115S	73-92	TNAU 71S	92-111
TNAU 116S	73-92	TNAU 101S	81-100
TNAU 124S	69-88	TNAU 135S	85-104
TNAU 137S	65-84	TNAU 136S	87-106
TNAU 139S	65-84	TNAU 147S	80-99
TNAU 142S	76-95	TNAU 151S	80-99
TNAU 143S	74-93		

It could be observed that five staggered sowings spread over a month had differential expression on days to heading among the TGMS lines studied. The difference was quite lower in some of the lines *viz.,* two days in TNAU 14S, three days in TNAU 18S, 71S and 116S and four days in TNAU 100S, 101S, 147S and 151S which is highly favourable during seed production since the time of sowing option is wider and its influence on flowering is less. In addition, these lines were completely sterile in all these sowings. On the other hand, TNAU 136S and 139S exhibited a wide flowering difference of 12 days in the same period followed by 10 days in TNAU 137S and nine days in TNAU 142S. These lines were also 100% sterile.

Pollen and spikelet fertility behavior in different Staggerings

Considering all the five staggerings, eight TGMS lines of medium duration category *viz.,* TNAU 46S, TNAU 67S, TNAU 100S, TNAU 116S, TNAU 124S, TNAU 137S, TNAU 139S and TNAU 143S and nine long duration genotypes *viz.,* TNAU 14S, TNAU 18S, TNAU 45S, TNAU 71S, TNAU 101S, TNAU 135S, TNAU 136S, TNAU 147S and TNAU 151S had shown 100% pollen and spikelet sterility (Table.4). Thus these lines have prolonged duration of sterility phase which is almost a month. Lu *et al*., (1998) [4] suggested that for successful utilization of TGMS lines, the sterile and fertile phase should be for minimum of 30 consecutive days in spite of the temperature changes during the sensitive stage. Latha and Thiyagarajan (2010) [2] reported six TGMS lines TS6, TS16, TS 18, TS 29, TS 46 and TS 47 that showed complete sterility for more than 50 consecutive days under high temperature (> 30/20°C) and TS 29 showed two distinct sterile phase one from mid February to mid June and other from second week of September to first week of November.

Table 4: Days to first flowering, critical stage and critical sterility point of 25 rice TGMS lines in weekly staggered sowings at Coimbatore

TGMS line	Date of sowing	Days to first flowering	Critical stage for sterility (days)	Minimum temperature (°C)	Maximum temperature (°C)	Pollen sterility (%)	Spikelet sterility (%)
TNAU 46S	02.01.2018	96	72-91	20.5	30.8	100.00	100.00
	10.01.2018	98	74-93	20.9	31.5	100.00	100.00
	17.01.2018	98	74-93	22.1	32.2	100.00	100.00
	24.01.2018	94	70-89	22.7	32.9	100.00	100.00
	31.01.2018	99	75-94	21.4	32.2	100.00	100.00
TNAU 67S	02.01.2018	104	80-99	21.0	31.6	100.00	100.00
	10.01.2018	102	78-97	21.5	31.8	100.00	100.00
	17.01.2018	101	77-96	22.7	32.9	100.00	100.00
	24.01.2018	102	78-97	21.8	32.8	100.00	100.00
	31.01.2018	106	82-101	21.8	32.8	100.00	100.00
TNAU 83S	02.01.2018	103	79-98	21.1	31.9	80.07	100.00
	10.01.2018	105	81-100	22.1	32.2	84.52	100.00
	17.01.2018	106	82-101	22.6	32.1	81.38	100.00
	24.01.2018	105	81-100	21.4	32.5	88.49	100.00
	31.01.2018	102	78-97	21.4	32.5	85.23	100.00
TNAU 84S	02.01.2018	96	72-91	20.5	30.8	94.74	100.00
	10.01.2018	98	74-93	20.9	31.5	97.05	100.00
	17.01.2018	99	75-94	22.3	32.4	99.50	100.00
	24.01.2018	96	72-91	22.9	33.0	98.30	100.00
	31.01.2018	97	73-92	22.9	33.0	99.20	100.00
TNAU 100S	02.01.2018	96	72-91	20.5	30.8	100.00	100.00
	10.01.2018	98	74-93	20.9	31.5	100.00	100.00
	17.01.2018	95	71-90	21.5	31.8	100.00	100.00
	24.01.2018	97	73-92	23.0	32.9	100.00	100.00
	31.01.2018	98	74-93	23.0	32.9	100.00	100.00
TNAU 114S	02.01.2018	97	73-92	20.6	30.9	39.47	70.81
	10.01.2018	101	77-96	21.2	31.8	35.60	78.60
	17.01.2018	104	80-99	23.0	32.9	42.58	81.50
	24.01.2018	103	79-98	21.5	32.8	28.52	83.50
	31.01.2018	102	78-97	21.5	32.8	33.48	81.42
TNAU 115S	02.01.2018	93	69-88	20.6	31.1	99.09	99.14
	10.01.2018	97	73-92	21.1	31.7	100.00	100.00

	17.01.2018	101	77-96	22.7	32.9	100.00	100.00
	24.01.2018	99	75-94	22.6	32.1	100.00	100.00
	31.01.2018	98	74-93	22.6	32.1	100.00	100.00
TNAU 116S	02.01.2018	97	73-92	20.6	30.9	100.00	100.00
	10.01.2018	99	75-94	20.9	31.6	100.00	100.00
	17.01.2018	97	73-92	21.9	32.0	100.00	100.00
	24.01.2018	97	73-92	23.0	32.9	100.00	100.00
	31.01.2018	98	74-93	23.0	32.9	100.00	100.00
TNAU 124S	02.01.2018	93	69-88	20.6	31.1	100.00	100.00
	10.01.2018	96	72-91	21.0	31.6	100.00	100.00
	17.01.2018	96	72-91	21.7	31.9	100.00	100.00
	24.01.2018	97	73-92	23.0	32.9	100.00	100.00
	31.01.2018	90	66.85	23.0	32.9	100.00	100.00
TNAU 137S	02.01.2018	89	65-84	20.3	30.4	100.00	100.00
	10.01.2018	94	70-89	20.8	32.2	100.00	100.00
	17.01.2018	93	69-88	21.0	31.7	100.00	100.00
	24.01.2018	98	74-93	22.8	32.5	100.00	100.00
	31.01.2018	99	75-94	21.4	32.2	100.00	100.00
TNAU 139S	02.01.2018	89	65-84	20.3	30.4	100.00	100.00
	10.01.2018	97	73-92	21.1	31.7	100.00	100.00
	17.01.2018	100	76-95	22.6	32.7	100.00	100.00
	24.01.2018	101	77-96	22.1	32.5	100.00	100.00
	31.01.2018	101	77-96	21.0	31.5	100.00	100.00
TNAU 142S	02.01.2018	94	70-89	20.6	31.0	99.69	100.00
	10.01.2018	98	74-93	20.9	31.5	99.10	100.00
	17.01.2018	103	79-98	22.9	33.0	100.00	100.00
	24.01.2018	100	76-95	22.4	32.1	100.00	100.00
	31.01.2018	102	78-97	22.8	32.5	100.00	100.00
TNAU 143S	02.01.2018	98	74-93	20.5	31.2	100.00	100.00
	10.01.2018	101	77-96	21.2	31.8	100.00	100.00
	17.01.2018	106	82-101	22.6	32.1	100.00	100.00
	24.01.2018	102	78-97	21.8	32.8	100.00	100.00
	31.01.2018	101	77-96	21.8	32.8	100.00	100.00
TNAU 145S	02.01.2018	96	72-91	20.5	30.8	92.68	89.50
	10.01.2018	100	76-95	21.0	31.7	93.58	98.40
	17.01.2018	101	77-96	22.7	32.9	91.23	95.48
	24.01.2018	97	73-92	23.0	32.9	95.87	95.60
	31.01.2018	100	76-95	23.0	32.9	92.48	93.93
TNAU 14S	02.01.2018	106	82-101	20.9	31.5	100.00	100.00
	10.01.2018	105	81-100	22.1	32.2	100.00	100.00
	17.01.2018	106	82-101	22.6	32.1	100.00	100.00
	24.01.2018	106	82-101	21.4	32.2	100.00	100.00
	31.01.2018	106	82-101	21.4	32.2	100.00	100.00
TNAU 18S	02.01.2018	108	84-103	21.0	31.7	100.00	100.00
	10.01.2018	109	85-104	22.8	33.2	100.00	100.00
	17.01.2018	110	86-105	21.5	32.8	100.00	100.00
	24.01.2018	109	85-104	21.0	31.2	100.00	100.00
	31.01.2018	110	86-105	21.0	31.5	100.00	100.00
TNAU 45S	02.01.2018	112	88-107	21.9	32.0	100.00	100.00
	10.01.2018	113	89-108	22.5	32.1	100.00	100.00
	17.01.2018	114	90-109	21.2	31.9	100.00	100.00
	24.01.2018	114	90-109	20.4	30.1	100.00	100.00
	31.01.2018	117	93-112	21.5	28.5	100.00	100.00
TNAU 71S	02.01.2018	114	90-109	22.3	32.4	100.00	100.00
	10.01.2018	116	92-111	21.8	32.8	100.00	100.00
	17.01.2018	114	90-109	21.2	31.9	100.00	100.00
	24.01.2018	115	91-110	20.2	29.9	100.00	100.00
	31.01.2018	116	92-111	21.4	28.5	100.00	100.00
TNAU86S	02.01.2018	106	82-101	20.9	31.5	48.79	65.14
	10.01.2018	107	83-102	22.6	32.7	43.70	72.60
	17.01.2018	105	81-100	22.8	32.5	52.80	68.90
	24.01.2018	106	82-101	21.4	32.2	46.10	75.20
	31.01.2018	105	81-100	21.4	32.2	58.30	78.30
TNAU 93S	02.01.2018	110	86-105	21.5	31.8	28.19	74.79
	10.01.2018	112	88-107	22.8	32.5	25.50	81.29
	17.01.2018	116	92-111	21.0	31.2	19.40	85.80
	24.01.2018	114	90-109	20.4	30.1	21.60	78.50
	31.01.2018	112	88-107	20.4	30.1	23.90	70.70
TNAU	02.01.2018	105	81-100	21.1	31.7	100.00	100.00

101S	10.01.2018	107	83-102	22.6	32.7	100.00	100.00
	17.01.2018	106	82-101	22.6	32.1	100.00	100.00
	24.01.2018	104	80-99	21.5	32.8	100.00	100.00
	31.01.2018	105	81-100	21.5	32.8	100.00	100.00
TNAU 135S	02.01.2018	109	85-104	21.2	31.8	100.00	100.00
	10.01.2018	108	84-103	22.7	32.9	100.00	100.00
	17.01.2018	106	82-101	22.6	32.1	100.00	100.00
	24.01.2018	109	85-104	21.0	31.2	100.00	100.00
	31.01.2018	104	80-99	21.0	31.5	100.00	100.00
TNAU 136S	02.01.2018	99	75-94	20.5	31.4	100.00	100.00
	10.01.2018	104	80-99	21.9	32.0	100.00	100.00
	17.01.2018	104	80-99	23.0	32.9	100.00	100.00
	24.01.2018	106	82-101	21.4	32.2	100.00	100.00
	31.01.2018	111	87-106	20.4	29.9	100.00	100.00
TNAU 147S	02.01.2018	106	82-101	20.9	31.5	100.00	100.00
	10.01.2018	104	80-99	21.9	32.0	100.00	100.00
	17.01.2018	106	82-101	22.6	32.1	100.00	100.00
	24.01.2018	107	83-102	21.2	31.9	100.00	100.00
	31.01.2018	104	80-99	20.0	28.0	100.00	100.00
TNAU 151S	02.01.2018	106	82-101	20.9	31.5	100.00	100.00
	10.01.2018	104	80-99	21.9	32.0	100.00	100.00
	17.01.2018	103	79-98	22.9	33.0	100.00	100.00
	24.01.2018	103	79-98	21.5	32.8	100.00	100.00
	31.01.2018	104	80-99	20.0	28.0	100.00	100.00

Figures in bold represent the minimum and maximum temperature (°C) for achieving the CSP for each TGMS line

In the remaining eight TGMS lines, the period of 100% sterility shows variations and also some lines did not record 100% sterility in any of the staggerings. The TGMS line TNAU 142S showed complete sterility for 14 days in the last three dates of staggered sowing (17th to 31st Jan) and TNAU 115S had shown sterility for 21 days except first staggering (10th to 31st Jan). Since these lines had very short sterility phase, they are unsuitable for commercial exploitation.

Two lines *viz.,* TNAU 83S and TNAU 84S exhibited complete spikelet sterility in all the five dates of staggered sowing but the former had 80.07 to 88.49% pollen sterility (partial sterility) and the latter had 94.74 to 99.50% pollen sterility. Chakrabarti *et al.,* (2010) [1] reported that the increased spikelet sterility than pollen sterility was due to reduction in the pollen germination. Four TGMS lines *viz.,* TNAU 86S, TNAU 93S, TNAU 114S and TNAU 145S had partial pollen and spikelet sterility in all the sowings of January and hence need to be evaluated in different month(s) to understand their sterility behaviour or otherwise.

Critical sterility point of TGMS lines

TGMS lines have complete pollen sterility at 30-34°C/22-23°C and partial to normal pollen sterility at 24-27°C/18-19°C (Sun *et al*., 1989; Virmani and Voc, 1991) [13, 16]. In our study, the lowest value of mean maximum temperature during the sensitive stage was considered as a parameter for determining the critical sterility point (CSP) of a TGMS line, since it is the maximum temperature that influences the fertility/sterility expression of the TGMS lines (Viraktamath and Virmani, 2001) [15] in tropical countries. Different TGMS lines had varying sensitive stages where temperature influences their sterility behavior.

The mean maximum and minimum temperatures during the critical stage for the TGMS lines exhibiting complete pollen and spikelet sterility for 30 days are shown in Fig.1. The lowest CSP of just 28°C was required for the lines TNAU 147S and TNAU 151S followed by 28.5°C for TNAU 45S and TNAU 71S. Sanchez and Virmani (2005) [9] emphasized the need for identification of TGMS lines with low critical sterility point because they remain completely sterile despite sudden temperature changes during the sensitive growth stage. The highest CSP of 31.7°C was required for TNAU 101S. Other lines that fall closer to these values were TNAU 67S (31.6°C), TNAU 14S, 18S and 135S (31.5°C). In earlier report by Sairekha *et al* (2018), the CSP of TNAU 45S was reported as 29.97°C, but in the present study, the line expressed complete sterility even at lower CSP of 28.5°C which is quite advantageous.

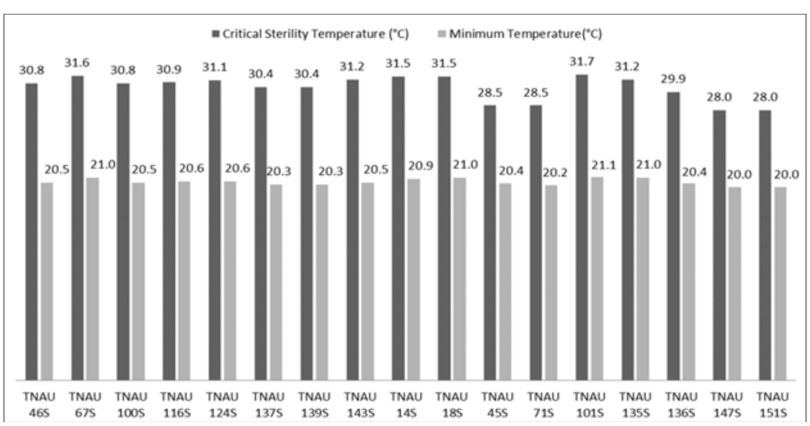

Fig 1: Maximum and minimum temperature (ºC) at 100% sterility for rice TGMS lines

The critical sterility temperature of TGMS lines studied ranged from 28.0 ºC to 31.7 ºC. Ali *et al* (1995) reported that in tropics, the ideal CSP is at maximum temperatures between 30 to 32 °C. The critical sterility point of six TGMS lines studied by Ramakrishna *et al.* (2006) ranged from 30ºC (DRR1S) to as high as 35.9°C (IR 73827-23S). Higher CSP from 33.9ºC to 35.8°C was reported by Salgotra *et al.* (2012) [8] for complete sterility of TGMS lines studied by them.

Lopez and Virmani (2000) [3] reported six new TGMS lines with complete pollen and spikelet sterility when the temperature was higher than 30 ºC. On the other hand, five lines *viz.,* Norin PL 12, ID 24, IR 32364 (TGMS), IR 72093 and IR 72096 were found to possess low CSP (T max <32°C) in the investigations by Sanchez and Virmani (2005) [9]

Influence of rainfall on temperature and sterility

Generally hybrid rice seed production using thermo-sensitive genic male sterility will be taken up during dry weather season so that the sensitive stage will coincide with high temperature and sterility is ensured. In our study, the flowering of TGMS lines coincided during hot months *viz.,* April and May during which the average maximum temperature was 32.0°C and 29.5°C respectively. The low temperature during May was due to the receipt of 162 mm rainfall. Such showers are expected during summer which poses a threat to TGMS hybrid seed production. A total of 145mm rainfall was received in five days (2.5.18 - 25mm, 5.5.18 - 48mm, 9.5.18 - 8.0mm, 10.5.18 - 52mm and 11.5.18 – 12mm) spread over ten days which coincided with the sensitive stage in the last staggering of 10 TGMS lines *viz.,* TNAU 67S, 14S, 18S, 45S, 71S, 101S, 135S, 136S,147S and 151S. The difference in mean maximum temperature between the fourth and fifth staggering was meager (0.0 to 0.3°C) in the lines TNAU 67S, 14S, 101S, 18S and 135S and moderate in TNAU 45S (1.6°C) and TNAU 71S (1.4°C). It was more pronounced in the lines TNAU 151S (4.8°C), TNAU 147S (3.9 °C) and TNAU 136S (2.3°C). Such a drop in temperature did not affect the sterility behavior of these lines. Incidentally, it helped in the identification of low CSP for the lines TNAU 45S (28.5°C), TNAU 71S (28.5°C), TNAU 136S (29.9 °C), TNAU 147S and TNAU 151S (28.0°C). All the above ten lines were 100% sterile in all staggerings.

Genotypic responses to temperature sensitivity and sterility

A study of pedigree of the lines taken for study reveals differences among the lines derived from same source for sterility behavior and CSP. The lines TNAU 142S, TNAU 143S and TNAU 145S were selections from TNAU 19S. The line TNAU 145S did not express complete sterility in any of the staggerings, whereas TNAU 142S was completely sterile in last three staggerings i.e. 15 days (17^{th} to 31^{st} Jan). The other line TNAU 143S was 100% sterile during the entire period of study. The lines TNAU 114S and TNAU 137S were derived from TNAU 4S but developed in crosses with different genetic background of BPT 5204 and CB 06564 respectively. Though the TGMS gene source was common, TNAU 114S was partially sterile while TNAU 137S was completely sterile. So also, TNAU 45S and TNAU 83S had common source but sterility differences were noticed. On the contrary, TNAU 67S and TNAU 135S possessing TS 29 source but developed from crosses with different male parents *viz.,* IR 62917-2-3-2R-1 and CO 49 respectively exhibited 100% sterility throughout the study period. The lines were of medium and long duration but their CSP was almost similar (31.6 and 31.2 °C).

Physical mutations of TS 29 resulted in the development of two TGMS lines *viz.,* TNAU 84S and TNAU 139S of medium duration. Their sterility behavior was altered in that, TNAU 139S was completely sterile in all staggerings but TNAU 84S did not exhibit 100% pollen sterility though spikelet sterility was 100%. Even at CSP of 33°C, TNAU 84S was only 99.2% sterile but CSP of TNAU 139S for complete sterility was low (30.4°C). Viraktamath and Virmani (2001) [15] studied TGMS lines IR 68945-4-33-4-14, IR 68949-11-5-31 and its *japonica* TMGS donor Norin PL12 for sterility induction. For inducing complete sterility more than 8 hours a day of exposure to temperature 32°C was essential in TGMS lines IR 68945-4-33-4-14 and IR 68949-11-5-31 but in case of Norin PL12, sterility was induced at only 4 hours a day exposure to the temperature of 32 °C. Thus it indicated that the changes in sterility expression in TGMS lines derived from same source may change due to the influence of different genetic background.

Conclusion

The result of this study showed that the among the 25 TGMS lines, 17 lines behaved as completely sterile for a period of 30 days. The sensitive stage for their sterility expression was 65 – 99 days for eight medium duration genotypes (TNAU 46S, TNAU 67S, TNAU 100S, TNAU 116S, TNAU 124S, TNAU 137S, TNAU 139Sand TNAU 143S) and 80-112 days for nine long duration genotypes (TNAU 14S, TNAU 18S, TNAU 45S, TNAU 71S, TNAU 101S, TNAU 135S, TNAU 136S, TNAU 147S and TNAU 151S). The critical sterility point ranged between 28°C to 31.7°C. Drop in temperature due to rainfall between the fourth and fifth staggering did not affect the sterility behavior in TGMS lines TNAU 151S (4.8°C), TNAU 147S (3.9°C) and TNAU 136S (2.3°C). Genotypic differences for expression of sterility existed even among the lines derived from same TGMS source. The study also resulted in the identification of low CSP lines which can be exploited on a commercial scale.

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