

Relative Efficiency of Different Emasculation Methods in Rice (*Oryza sativa* L.)

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Abstract The efficiency of an emasculation method based on amount of self seed set determines the trueness of hybrids and quantity of hybrid seed produced. Quantitative comparisons between emasculation techniques in rice are scarce and the objective of present investigation was to assess the relative efficiency of four different emasculation methods viz. hand emasculation, suction method, 70% ethanol and absolute ethanol based on the amount of self seed set upon emasculation followed by no pollination. The comparison of mean efficiency of emasculation methods over five genotypes and two seasons revealed that hand emasculation (84.05% efficiency) followed by suction method (75.74% efficiency) were most efficient, that is least self seeds were set upon emasculation followed by no pollination. The

study also revealed that there were no genotypic differences for emasculation efficiency. Keeping in view the results, we recommend hand emasculation the most efficient but laborious method. However, suction method will be very useful as time saving and less laborious method for removal of anthers in rice.

Keywords Rice, Emasculation, Self seed, Efficiency.

Introduction

Emasculation is an essential pre-requisite for undertaking hybridization between two or more crop plants. In order to successfully cross-pollinate two different rice plants, the anthers must be removed from the rice floret so that self-pollination will not occur [1]. The hybrid seed produced may be either supplied to farmers for general cultivation or may be used for evaluation of combining ability among different rice genotypes. several types of emasculation methods are available in rice viz., hand emasculation, suction method, alcohol treatment, hot water and cold water emasculation. At International Rice Research Institute (IRRI), emasculation is made by anther aspiration [2]. They got 40—50% hybrid F1

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plants in the dry season, 20—30% in the wet season. This indicates that efficiency of method of emasculating in rice varies with season/environment.

The efficiency of any of these emasculating methods determines the trueness of the hybrids and the hybrid seed set upon crossing. Hence, determination of an efficient emasculating method is essential for sexual hybridization-mediated gene transfer and hybrid development based crop improvement programs. The utilization of the determined efficient emasculating method will allow us to achieve the desired goals in crop improvement programs much precisely. Keeping in view, the varied efficiency of emasculating methods over environments/locations and scarce quantitative comparisons among emasculating methods in rice, the present investigation was carried out to compare the efficiency of different emasculating methods during two seasons (*kharif* 2014-15 and 2015-16) at crop Research Center of G B Pant University of Agriculture and Technology, Pantnagar, Uttarakhand, based on the amount of self seed set in emasculated panicles without pollination.

Materials and Methods

The present investigation was used to compare the efficiency of four emasculating methods viz., hand emasculating, suction method and alcohol treatment (70% and absolute ethanol). Ten panicles of each of the five genotypes (TN1, Pusa Basmati 1, MRST 3, PHS 29 and MRST 7) were subjected to each of the four emasculating methods during each season. In all the methods, mother plants were subjected to emasculating when one third to one half of the panicle was exerted. Self-pollination can occur by emasculating florets which are too mature. In order to minimize this risk, emasculating was effected in florets at the middle part of the panicles, while florets at the top bottom were discarded in all the four techniques. All the four methods were practiced during evening hours starting from 4 PM. In case of hand emasculating, the anthers were removed from florets in middle part of the panicle with the help of forceps without injuring the stigma. Whereas, in case of suction method the anthers were removed with a microtip connected by a plastic tube to a motor which

acts as vacuum aspirator [3, 4]. As far as alcohol treatment is concerned, the panicles (with top and bottom florets clipped) were dipped for two minutes in a beaker carrying 70% ethanol and 100% ethanol. The panicles subjected to the emasculating methods were bagged with a butter paper bag and labeled appropriately. The data was recorded on the number of self seeds set (without pollination) per 100 florets emasculated. The efficiency of emasculating method was worked out as per the formula given below :

Efficiency of emasculating (e)

$$= \left(1 - \frac{\text{Number of selfed seeds}}{\text{Total number of florets subjected to given emasculating method}} \right) \times 100$$

The percent values (Proportions) of efficiency of each emasculating technique averaged over all the genotypes during each season were statistically compared using Z-test of significance [5] and the Z values were calculated using Microsoft Office Excel. The emasculating efficiency averaged over the season and genotypes of each method was also compared for statistical significance using Z-test.

Results and Discussion

Emasculating being an important pre-requisite for sexual gene transfer across crop plants. However, the efficiency of an emasculating method is very important for production of true F_1 seeds, that too in abundant quantity. In rice, several methods are available for emasculating the florets. The present investigation was carried out to assess the relative efficiency of four types of emasculating methods in rice viz., hand emasculating, suction method, 70% and absolute ethanol treatment on the basis of amount of self seed set.

The investigation revealed significant differences among emasculating methods for percent mean efficiency over five genotypes during each season. During *kharif* 2014-15, the percent efficiency of hand emasculating method ranged from

Table 1. Efficiency of various emasculations methods exercised on five different rice genotypes during *kharif* 2014-15. f = number of florets emasculated; s = self seeds obtained from emasculated florets without pollination; e = efficiency of emasculations method; *Significant differences at $p > 0.05$ level of significance (evaluated using Z test of significance).

Genotypes	Hand emasculations			Suction method			70% ethanol			100% ethanol		
	f	s	e	f	s	e	f	s	e	f	s	e
TN 1	300	40	86.67	400	84	79.00	370	109	70.54	280	92	67.14
Pusa Basmati 1	450	57	87.33	460	101	78.04	350	95	72.86	360	111	69.17
MRST 3	280	55	80.36	380	69	81.84	302	120	60.26	375	102	72.80
PHS 29	350	52	85.14	305	88	71.15	370	110	70.27	410	105	74.39
MRST 7	310	53	82.90	330	89	73.03	410	105	74.39	305	107	64.92
Mean	338	51.4	84.48*	375	86.20*	76.61	360.4	107.8	69.66	346	103.4	69.68

82.90 to 87.33 with mean efficiency of 84.48%. As far as suction method is concerned, the percent efficiency ranged from 71.15 to 81.84. Whereas, the range was 60.26 to 74.39% and 64.92 to 74.39% in case of 70% and absolute ethanol based emasculations, respectively. The percent mean efficiency over all the genotypes obtained through suction method, 70% ethanol and absolute ethanol was 76.61, 69.66 and 69.68 respectively (Table 1). During *kharif* 2015-16, the mean efficiency of hand emasculations, suction method, 70% and 100% ethanol treatments was 83.63, 74.87, 66.60 and 66.30%, respectively (Table 2). The mean efficiency of hand emasculations over the two seasons of hand emasculations was 84.05%. Whereas, it was 75.74, 68.13 and 67.09% for suction method, 70% and 100% ethanol treatment, respectively (Table 3). As far as influence of genotypes over efficiency of an emasculations method are concerned, no significant differences were identified among genotypes. This infers that emasculations efficiency is genotype independent and it only depends on the physical tech-

nique of removing or killing anthers. The percent mean emasculations efficiency over all the emasculations techniques and seasons of genotypes TN 1, Pusa Basmati 1, MRST 3, PHS 29 and MRST 7 was 74.42, 74.86, 73.98, 74.71 and 71.94 (Table 1).

The above data reveals that hand emasculations method was significantly superior in percent mean efficiency as compared to other methods, that is, least self seeds were produced in panicles subjected to hand emasculations followed by no pollination. Garcia-Yzaguirre and Carreres claimed that hand emasculations followed by brush pollination of crushed anthers produced an average of 48.2% hybrid F_1 plants [6]. The next best emasculations method in terms of superiority in less self seed set, was suction method which was significantly superior to 70% and absolute ethanol based emasculations. Berdrejo [7] and Sha [8] also recommended hand emasculations as the most efficient but laborious technique and suggested utilization of alcohol based emasculations under circumstances of labor shortage. Re-

Table 2. Efficiency of various emasculations methods exercised on five different rice genotypes during *kharif* 2015-16. f = number of florets emasculated; s = self seeds obtained from emasculated florets without pollination; e = efficiency of emasculations method; * Significant differences at $p > 0.05$ level of significance (evaluated using Z test of significance).

Geno- type	Hand emasculations			Suction method			70% ethanol			100% ethanol		
	f	s	e	f	s	e	f	s	e	f	s	e
TN 1	255	37	85.49	258	63	75.581	252	88	65.08	240	82	65.83
Pusa Basmati 1	240	47	80.42	202	53	73.76	237	73	69.20	310	99	68.06
MRST 3	344	51	85.17	199	56	71.86	201	55	72.64	275	91	66.91
PHS 29	277	47	83.03	255	54	78.82	242	64	73.55	248	96	61.29
MRST 7	263	42	84.03	230	59	74.35	198	94	52.53	330	101	69.39
Mean	275.8	44.8	83.63*	228.80	57.00	74.87*	226.00	74.80	66.60	280.60	93.80	66.30

Table 3. Mean efficiency of various emasculaton methods exercised on five different rice genotypes over two the years.* Significant differences at $p>0.05$ level of significance (evaluated using Z test of significance).

Geno- type	Efficiency (%)				Mean
	Hand emas- culation	Suction method	100% ethanol	70% ethanol	
TN 1	86.08	77.29	67.81	66.49	74.42
Pusa Basmati 1	83.88	75.90	71.03	68.62	74.86
MRST 3	82.77	76.85	66.45	69.85	73.98
PHS 29	84.09	74.99	71.91	67.84	74.71
MRST 7	83.47	73.69	63.46	67.16	71.94
Mean	84.05*	75.74*	68.13	67.99	

searchers at IRRI reported that emasculaton using immersion methods (alcohol immersion or hot water immersion) had lower seed set than hand or vacuum (suction) emasculaton because culms were bent to introduce panicles into a container which affects seed set and may result in breakage of panicles [9].

Conclusively, it can be stated that hand emasculaton though being most efficient in terms of the lowest self seed set , yet, it is time consuming as compared to as other emasculaton methods, thereby,

incurring extra costs and labor. Suction method although being less efficient as compared to hand emasculaton, however, it is less time consuming and hence can be more useful in rice hybridization programs.

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