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Inheritance Studies for Stigma exsertion in F₃ Population of Rice Maintainer Lines

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Abstract

Stigma exsertion is important in increasing out crossing rate in rice hybrid parental lines. In the present investigation, inheritance pattern of stigma exsertion trait in F_3 population developed between IR68897B x BF16B was studied. On phenotyping of 674 F_3 plants, it segregated into 91 low, 456 moderate and 127 high stigma exsertion plants based on IRRI-SES scale published by INGER, 2014. Further, subjected to chi square analysis to test their goodness of fit to appropriate Mendelian ratios and showed significant. Thus, stigma exsertion is quantitative trait. Also transgressive segregation observed during phenotyping indicated that complementary gene action with additive effects and dispersed in both the parents. The frequency distribution showed continuous variation signifying that stigma exsertion trait is controlled by polygenes and influenced by environment.

Key words: Inheritance, polygenes, F₃ population, maintainer line.

Introduction

Asia produces and consumes 90% of the world's rice (FAO Rice Market Monitor, 2015). By 2030, world must produce 70% more rice than it produced in 1995 to meet demand of increasing population and rising incomes (Yuan, 2005). This production increase must be achieved with less and degraded land, less labour, less water and less environment pollution and it must be sustainable in changing climate. Hybrid rice cultivation is a tool among available technologies to increase yield and food security (Virmani and Kumar, 2004).

In china, rice hybrids are grown in large acreage since 1974. Use of hybrid rice increases yield by 20-25% compared to inbred lines/high yielding varieties (Yuan, 2005). After the success of China's demonstration of rice hybrid cultivation, hybrid rice cultivation has been started in India, but, not on a large scale due to various reasons. Among them, high seed cost is major one. Rice is self pollinating crop and securing natural out-crossing on male sterile plant is gaining importance to reduce cost of seed production (Virmani 1994). The out-crossing can be improved through GA3 application and depends on many floral traits *viz.*, number of opened spikelets, duration and angle of spikelet opening, pollen load, pollen longevity, length of style, panicle exsertion, flag leaf angle and length

and stigma characters (length, width and receptivity). Among them, consistent importance is given by rice researchers for stigma traits since it gives opportunities to increase the out crossing percentage (Kato *et al.*, 1987). Also different types of stigma viz., trifid, dual, single types directly influence seed production efficiency (Virmani and Athwal, 1973; Yan *et al.*, 2009).

Earlier studies suggest that, stigma exsertion is partially dominant (Virmani and Athwal, 1974) or completely dominant (Li *et al.*, 1979, 1981) Li *et al.* (1985) reported that, stigma exsertion is a quantitative character. This study was conducted to know inheritance of stigma exsertion traits using rice maintainer line F_3 population.

Materials and Methods

Study materials developed at ICAR-Indian Institute of Rice Research (IIRR), Hyderabad, Telangana. Details of materials used in conduct of experiment and statistical analysis of data are presented.

Plant Materials

IR68897B, a maintainer line of early hybrid DRRH-2, with low stigma exsertion (36.78%) and BF-16B, maintainer line with high stigma exsertion (80.25%) were chosen as parents for development of F_3 mapping population. Hybrid nature of F_1 s was confirmed using microsatellite (SSR) markers



that were polymorphic between the parents. F_1 plants were selfed to generate F_2 population and F_2 selfed to generate F_3 population during *kharif* 2014. Panicles were collected at the time of post anthesis. For phenotypic evaluation of stigma exsertion trait, 674 F_3 plants were grown along with parents and F_1 s under field conditions during *rabi* 2014.

Phenotypic evaluation

For assessing stigma exsertion type, whole panicle method as described by Akhilesh *et al.*, 2015 was followed. All the individual spikelets in each panicle were separated and observed using magnifier lens to categorize them into dual, single or no stigma exsertion types (Fig. 1).



Figure 1: Stigma exsertion types (dual, single and no SE types)

Spikelets representing each class of stigma exsertion were counted separately and represented as percentage, as detailed by Yan *et al.* (2009).

Single stigma exsertion [SStgE] (%) = [SStgE / (SStgE + DStgE + NStgE)] x100

Dual stigma exsertion [DStgE] (%) = [DStgE/ (SStgE +DStgE +NStgE) x100

Total stigma exsertion [TStgE] (%) = SStgE (%) + DStgE (%) and

No stigma exsertion [NStgE] (%) =100 - TStgE (%).

Frequency distribution of stigma exsertion in F_3 population was calculated, as per the scale given (in the following table) by IRRI (SES, 2014).

Table 1: Stigma exsertion rate scale given by IRRI(SES, 2014)

Scale	Stigma exsertion rate (%)	Classification	
1	Above 70	High	
3	40-69	Medium/Moderate	
5	21-39		
7	11-20	Low	
9	0-10		

Phenotypic data analysis

Chi-square value was calculated using following formula.

$$\chi^2 = \frac{\sum (O-E)^2}{E}$$

Where, O = Observed frequency

E = Expected frequency

 Σ = Summation of the data

If the calculated values of χ^2 was high at 5 per cent level of significance, is said observed frequencies are not in accordance with the hypotheses and *vice versa*.

Result and Discussion

Stigma exsertion (SE) is an important floral character for improving hybrid seed production. Knowledge of genetics of a trait helps to effectively select the parents and choose an appropriate breeding method (Sarker *et al.*, 1999). Breeding for well exserted stigmas is one of the main approaches to solve the problem of low seed yield in hybrid seed production.

In this study, inheritance of stigma exsertion trait in F_3 population of maintainer lines of hybrid rice is studied. For phenotypic evaluation of stigma exsertion trait, 674 F_3 plants derived from IR68897B and BF-16B, a low (36.78%) and high (80.25%) stigma exsertion maintainer lines, respectively were grown along with parents and F_1 .s under field conditions. Hybridity of F_1 was confirmed using microsatellite (SSR) markers that were polymorphic between the parents (Fig. 2). F_1 plants were selfed to generate F_2 and F_2 were selfed to generate F_3 population. While, F_1 was lower than better parent and higher than recipient parent *i.e.*, intermediate with 60.44% stigma exsertion rate. This would be expected if multiple factors with cumulative effect governed this trait.

The F_3 population subjected to phenotyping by whole panicle method described by Akhilesh *et al* 2015.

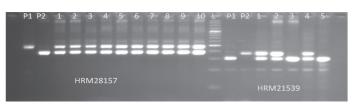


Figure 2: Confirmation of F1 with HRM 28157 and HRM 21539



SE percent for each panicle was calculated and F_3 population classified into different classes. The phenotyping classes were subjected to χ^2 analysis to test their goodness of fit to appropriate Mendelian ratios (Table 2).

Table 2: Segregation pattern of stigma exsertion trait in the F_3 population between IR-68897B and BF-16B.

Segregation	Observed	Expected	χ2	χ^2 table
for SE	frequencies	frequencies	value	value (2 df)
Low stigma	105	169	24.24	
exsertion				
Midium	442	337	32.72	
stigma				
exsertion				
High stigma	127	168	10.01	
exsertion				
Total	674	674	66.97*	5.99

SE-Stigma exsertion, *significant at 5% level

The F₃ population segregated in the ratio of 0.62:1.31:0.75 (105 low: 442 Moderate: 127 High) for stigma exsertion. Table value of χ^2 at 0.05 level of probability at 2 df was 5.99. The calculated χ^2 value (66.97), was more than the table χ^2 value at 2 df (5.99). χ^2 value was significant thus, stigma exsertion trait is governed by polygenes.

Frequency distribution of stigma exsertion

The frequency distribution of stigma exsertion rate in F_3 population showed a continuous variation (Table 3). The total phenotypic data was divided as per the scale of IRRI-SES system by INGER (SES, 2014).

Table 3: Frequency distribution of stigma exsertion in F_3 population

Stigma exsertion %	No. of plants	
0-10%	1	
11-20%	8	
21-40%	96	
41-70%	442	
>70%	127	
Total	674	

The graph of percentage of stigma exsertion (X-axis) plotted against number of plants (Y-axis) showed a bell shaped normal distribution curve and skewed towards low stigma exsertion. The frequency distribution of stigma exsertion (Fig. 3) indicated that both parents have several chromosomal regions increasing the frequency of stigma exsertion and trait is affected by environment.

In this study, transgressive segregants were observed in F_3 population (Table 2). Occurrence of transgressive

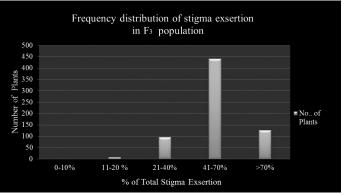


Figure 3: Frequency distribution of stigma exsertion of F_3 population between IR-68897B and BF-16B

segregants signifies complementary action of the genes with additive effect that had been dispersed in both the parents. According to Singh and Narayanan (1993) transgressive segregations are only possible from crosses between two parents with contributing alleles for quantitative trait and such segregants are not possible in case of qualitative traits. De Vicente and Tanksley (1993) studied that, transgressive segregation is commonly observed in the population segregating for quantitative trait; whereas Ricks and Smith (1953) mentioned that, transgressive segregation in certain progenies is because of accumulation of complementary alleles at multiple loci inherited from both parents.

The results obtained in the present study were in conformity with the results of Virmani and Athwal (1974), Li and Chen (1985), Wang et al. (2008), Ling (1989), Lou et al. (2014) and Pingbo et al. (2014) who observed continuous variation and studied genetics of the trait with conclusion that stigma exsertion trait of rice is a quantitative trait controlled by polygenes. However, some earlier reports on genetics of stigma exsertion in rice showed contradiction to the present finding. Hassan and Siddiq (1984) reported that fully exserted stigma is monogenic and dominant over partially exserted stigma exsertion. Xu and Shen (1987) also studied inheritance of stigma exsertion trait and reported that, it was a dominant trait. Experimental evidence suggested that environmental effects would also produce a continuous variation even if the number of genes governing a character was very small or even one.

Stigma exsertion is considered as an important floral characteristic for promoting out crossing rate especially in self-pollinated crops. In this study, we confirm quantitative inheritance of stigma exsertion in rice that would help in increasing hybrid seed production.



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