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Genetic studies on common rust (*Puccinia sorghii*) of maize under Kashmir conditions

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Common rust caused by fungus *Puccinia sorghii* is among one of the worldwide spread foliar corn diseases. The disease has persisted with varying degrees of susceptibility on higher altitudes in Kashmir valley. Disease reaction studies against common rust were carried out in two crosses involving indigenously identified cytosterile source viz.,15C (A) x I-318 (R) and I-401(A) x I-318(R) for all the six basic generations with P_1, P_2 and F_1 having 30 plants each and F_2 (300 plants), BC_1 (180 plants) and BC_2 (180 plants). Analysis of variance of arc sin transformed data in the present study revealed significant differences among all the generations of both the crosses suggesting presence of sufficient variability for prevalence of diseases. Common rust screening indicated the presence of resistant genes in both the crosses which further can be exploited in the production of successful commercial hybrids by using these cytoplasmic male sterility (CMS) sources as parents to develop *Puccinia sorghii* resistant, cost effective and stable hybrids.

Key words: Maize, common rust, cytosterile source.

INTRODUCTION

Across the globe today, maize (*Zea mays* L.) is a direct staple food for millions of people and through indirect consumption as a feed crop, is an essential component of global food security. Maize is produced on nearly 100 million hectares in developing countries, with almost 70% of the total maize production in the developing world coming from low and lower middle income countries (FAOSTAT, 2010). By 2050, demand for maize will double in the developing world, while by 2025 maize is predicted to become the crop with the greatest production globally (Rosegrant *et al*., 2008).

In the state Jammu and Kashmir, maize is second most important crop after rice and is a staple food of some tribal areas such as Gujar and Bakarwall (nomadic race).

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The main maize crop is generally grown as rainfed and on marginal lands particularly in hilly terrains of the Kashmir valley. In Kashmir valley the crop is cultivated on an area of 0.1 million ha area with production of 0.15 million tonnes with an average yield of 1.2 tonnes ha^{-1} (Anonymous, 2012). This low productivity is due to both biotic and abiotic stresses and among them common rust is a problem in late planted maize grown on higher altitudes of valley and therefore should receive high priority in maize breeding research. Common rust of maize caused by *Puccinia sorghi* (Schw.) caused a yield loss of 12-60% (Utpal et al., 2012) and occurs more in spring maize growing areas (Wang et al., 2014). Very little work has been carried out in Kashmir valley on common rust due to its low prevalence in plains but on higher altitudes the disease has persisted with varying degrees of susceptibility. *Puccinia sorghi* was first described by Schweinitz from plant tissue he thought to be sorghum. While the species name would suggest sorghum is a host, *P. sorghi* does not infect Sorghum spp. The uredial and telial hosts are corn, annual teosinte (*Euchlaena* (*Zea*) *Mexicana* Schrad.), and perennial teosinte (*E.* (*Zea*) *perennis* Hitch.) (Ullstrup, 1977). Common rust may cause extensive yellowing and premature desiccation of maize foliage, resulting in leaf necrosis, and complete destruction of photosynthetic areas. In extreme cases, heavy rust infestations may result in stunting, incomplete ear tip fill and pustules on ear husks, reducing marketability and yield. Common rust of maize may be controlled by partial or hypersensitive resistance. More than 20 hypersensitive resistance *(Rp)* genes have been identified against common rust in corn germplasm (Hooker, 1969).

Thus, hybrids with resilience to *P. sorghii* provide an effective way of achieving higher production and productivity levels of the crop. But considering the cost factor cytoplasmic male sterility (CMS-source) provides a sound and sustainable alternative besides adding purity to the end product. Therefore, in the present study screening for common rust was carried out in all the six generations of both the crosses viz., 15C (A) x I-318 (R) and I-401 (A) x I-318 (R) (Where A is Female MS; Ris Male fertile parent).

MATERIALS AND METHODS

The material for study was developed by attempting the crosses 15C (A) x I-318 (R) and I-401 (A) x I-318 (R) during *Kharif* (Summer season) 2010 to generate F_1 generation at High Altitude Rice Research Sub-station, Larnoo. The F_2 and backcrosses generation (BC_1 and BC_2) were developed at Winter Nursery Centre (ICAR) Hyderabad, during *Rabi* 2010-11. All the six basic set of generations P_1, P_2, F_1, F_2, BC_1 and BC_2 and 15C (B), I-401 (B) and restorer R-line I-318 (R) of the crosses thus obtained were raised and screened for common rust.

Six generations of each cross were evaluated in randomized complete block design with three replications at the Experimental Farm of Division of Plant Breeding & Genetics, SKUAST-K, Shalimar during *Kharief* 2011. The non-segregating (P_1, P_2 and F_1) and segregating generations (F_2, BC_1 and BC_2) were raised in four and six rows with inter and intra row spacing of 60 cm and 25 cm, respectively. Screening for disease was carried out with 30 plants each of P_1, P_2 and F_1 and 300 plants of F_2, 180 plants each from BC_1 and BC_2.

Inoculum for trial was prepared from uredospores collected from corn leaves from different leaves in Kashmir valley, sealed in glass vials and stored at 21°C. Percentage of uredospores germinated was determined on 2% water agar. Uredinospore concentration was adjusted so that about 0.125 g of viable uredinospores per litre of water containing two drops of Tween-20 as wetting agent. Inoculations were made when plants were approximately at mid silk stage of growth. Rust ratios were taken 18 days after inoculation on all plants.

Assessment was done following the scale described by Zummo (1988) which is a modification and expansion of scale devised by Ullstrup (1945) as: 0=Absence of infection (no pustules); 1=slight infection (a few isolated pustules); 2=light infection (prominent pustules not so scattered); 3=moderate infection (upto 5 % of leaf area infected); 4=heavy infection (6-15% of leaf area infected); 5=very heavy infection (16% or more of leaf area infected).

The per cent disease incidence and severity were calculated at each observation as per the following formula: per cent disease incidence = number of diseased leaves / total number of leaves assessed x 100 and per cent disease severity = sum of all numerical ratings / number of leaves examined x maximum disease rating x 100. The data was arc sine transformed as recommended for data, expressed as decimal fractions or percentages as per the procedure of Steel and Torrie (1980).

RESULTS AND DISCUSSION

The observations on the evaluation of genotypes in all six generations P_1, P_2, F_1, F_2, BC_1 and BC_2 of two crosses viz., 15C (A) x I-318 (R) and I-401 (A) x I-318 (R) against *Pucciniasorghii* were recorded by calculating per cent disease incidence and per cent disease severity following (0-5) scale of Zummo (1988) respectively. The data was arc sine transformed as per Steel and Torrie (1980) and analysis of variance for the transformed data for both the crosses is presented in Table 1. Results reveal significant differences among generations of both crosses suggesting presence of sufficient variability for prevalence of diseases.

Significant critical difference of 2.06 and 2.27 with respect to rust incidence and significant critical difference of 1.96 and 1.58 with respect to rust severity were observed in cross 15C (A) x I-318 (R) and I-401 (A) x I-318 (R). Responses of the two crosses viz., 15C (A) x I-318 (R) and I-401 (A) x I-318 (R) to *Pucciniasorghii* in the trial (*Kharief* 2011) are shown in Figure 1.

Subsequently six generations of the two crosses were grouped into moderately resistant (MR) category for common rust based on the severity of disease. The crosses15C (A) x I-318 (R) and I-401 (A) x I-318 (R) were moderately resistant (MR) to common rust with mean disease severity ranging from (9.44-16.52) in 15C (A) x I-318 (R) and (9.42-17.64) in I-401 (A) x I-318 (R) cross as revealed in Table 2.

Analysis of variance detected significant differences between generations of both crosses indicating sufficient

Table 1. Analysis of variance of arc-sine transformed generation means for reaction to common rust (*Puccinia sorghii*) in two crosses I-15(A) x I-318(R) and I-401(A) x I-318(R) of maize.

Cross : I-15C(A) x I-318(R) [Rust incidence]						**Cross : I-401(A) x I-318(R) [Rust incidence]**					
S.V	**D.F**	**S.S**	**M.S**	**F**	**P**	**S.V**	**D.F**	**S.S**	**M.S**	**F**	**P**
Rep.	3	21.148	7.049	0.88	0.476	Rep.	3	6.76	2.25	0.22	0.88
Treat.	5	379.64	75.929	9.43	0.00	Treat.	5	562.02	112.40	10.82	0.000
Error	15	120.76	8.051			Error	15	155.88	10.34		
Total	23	521.56				Total	23	724			
$S.E_{(diff.)}$ = 2.006; C.D = 4.27**						$S.E_{(diff.)}$ = 2.27; C.D = 4.83**					
Cross : I-15C(A) x I-318(R) [Rust severity]						**Cross : I-401(A) x I-318(R) [Rust severity]**					
S.V	D.F	S.S	M.S	F	P	S.V	D.F	S.S	M.S	F	P
Rep.	3	0.852	0.284	0.547	0.672	Rep.	3	12.397	4.13	4.30	0.26
Treat.	5	96.506	19.301	37.18	0.000	Treat.	5	87.88	17.57	19.08	0.000
Error	15	7.792	0.519			Error	15	14.40	0.960		
Total	23	105.15				Total	23	114.677			
$S.E_{(diff.)}$ = 0.597; C.D = 1.196**						$S.E_{(diff.)}$ = 0.647; C.D = 1.588**					

** = Significant at 5% level.

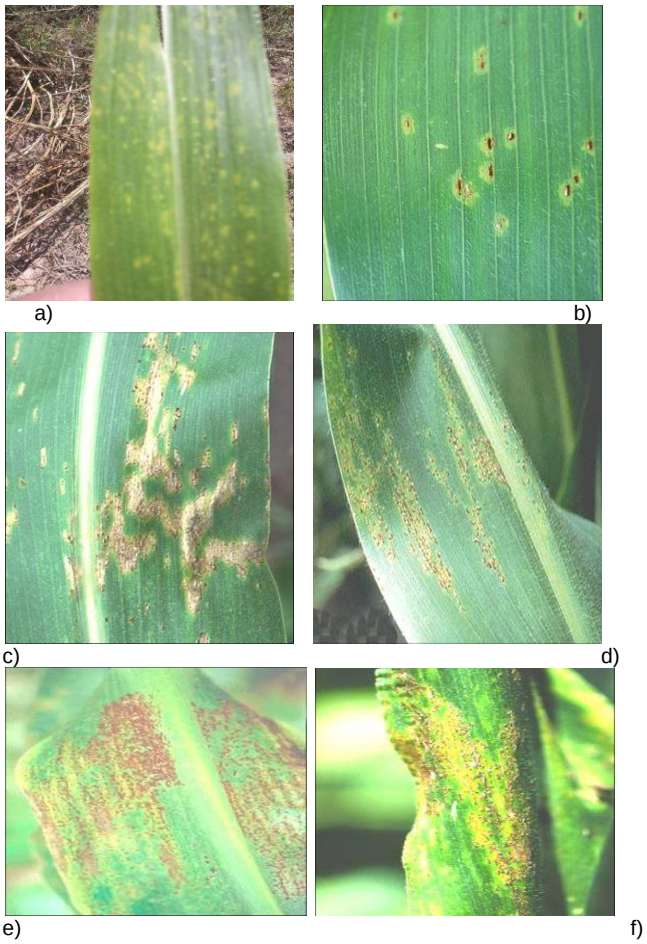

Figure 1. Common rust symptoms in two crosses viz. I-401(A) xI-318 (R) and I-15C(A) x I-318 (R). **a)** Initial symptom of leaf rust; **b)** prominent rust spores; c) ruptured rust Pustules**; d)** small amount of pustule development on leaf; **e)** uredospores of common rust; **f)** shriveled leaf after rupture of rust spores.

Table 2. Arc sine transformed mean disease incidence and severities of six generation of six generations of two crosses 15C(A) x I-318(R) and I-401(A) x I-318 (R) of maize for reaction to common rust (*Puccinia sorghii*).

Parameter		Mean disease incidence (%)	Mean disease severity (%)	Reaction
Cross 15C(A) x I-318(R)				
P_1	I-15C(A)	27.73 (31.72)	8.09 (16.52)	MR
P_2	I-318(R)	14.75 (22.52)	3.30 (9.42)	R
F_1	I-15C(A) x I-318(R)	11.06 (19.12)	2.94 (9.6)	R
F_2	I-15C(A) x I-318(R)	16.62 (24.00)	3.67 (11.03)	R
BC_1	[I-15C(A) x I-318(R)] I-15C(A)	17.54 (24.68)	4.78 (12.53)	MR
BC_2	[I-15C(A) x I-318(R)] I-318(R)	12.91 (20.94)	2.75 (9.44)	R
Cross I-401(A) x I-318(R)				
P_1	I-401(A)	31.44 (34.09)	9.20 (17.64)	MR
P_2	I-318(R)	14.75 (22.52)	3.30 (9.42)	R
F_1	I-401(A) x I-318(R)	11.07 (19.36)	5.51 (13.55)	MR
F_2	I-401(A) x I-318(R)	14.15 (22.07)	7.18 (15.49)	MR
BC_1	[I-401(A) x I-318(R)] x I-401(A)	20.32 (26.55)	7.73 (16.08)	MR
BC_2	[I-401(A) x I-318(R)] x I-318(R)	13.85 (21.64)	4.59 (12.32)	R

Source: Ullstrup (1945), Zummo (1988). Resistant (<5% of leaf area infected); MR = Moderately resistant (5.0-25.0% of leaf area infected).

variability for disease infestation but environmental factors particularly high temperature in plains might have been the limiting factor in development of maize rust. Since all the generations of both studies crosses were categorized into one group that shows moderate resistance. Little rust development in both crosses (Figure 1) reveal presence of some dominant gene which might have passed from parents. Although there was variation in disease severity between the crosses but in general all the generations of the studied crosses had fewer pustules which were significantly smaller. The percentage of pustules that had ruptured was less indicating that latent period was somewhat longer in these crosses suggesting presence of resistant genes which might be responsible for slow rusting characteristics in these crosses. Presence of slow rusting characteristics in the studied crosses reduced number and size of pustules (Figure 1), limiting secondary inoculums and thus very less plant damage was visible.

In some studies, more than 25 dominant resistance (*Rp*) genes were found to be involved in race specific resistance and organized in complex loci at chromosomes 3, 4, and 10 (Hooker, 1985; Delaney et al., 1998). Richter et al. (1995) found that within these complex loci, novel resistance specificities are generated by genetic re-assortment events, such as unequal crossing over or gene conversion. Pyramiding of multiple closely linked genes into “compound” genes has been proposed as a possible means of constructing more durable race specific resistance inherited by complex loci against common rust in maize (Hu and Hulbert, 1996). Here in this study, the results indicated presence of resistant genes in both crosses which can be further exploited in the production of successful commercial hybrids by using these CMS sources as parents to develop *P. sorghii* resistant, cost effective and stable hybrids. Also further testing of these crosses for rust resistance through molecular markers can be helpful in identifying resistant gene in commercial hybrids.

Conflict of interests

The authors did not declare any conflict of interest.

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