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Short Communication

New sources of resistance to spot blotch in emmer wheat developed through mutagenesis

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In India, three species of wheat *viz*., bread wheat (*Triticum aestivum* L.), macaroni wheat (*Triticum durum* Desf.) and emmer or dicoccum wheat (*Triticum dicoccum* (Schrank.) Schulb.) are cultivated. Scientific studies related to dicoccum wheat revealed that they are nutritionally superior as compared to commercially available bread and durum wheat with high protein and dietary fiber contents. Dicoccum based products have high satiety value, low digestibility, low glycemic value and has been considered as a therapeutic food in the management of diabetes [1]. Emmer wheat is endowed with natural resistance to brown and black rusts but highly susceptible to spot blotch caused by *Bipolaris sorokiniana* Shoem. (*Cochliobolus sativus* Drechs. ex Dastur), which is recently a major concern in Mega Environment 5 (ME 5) characterized by a warm humid climate [2]. The spot blotch disease is recently gaining much importance in Karnataka State of India because of occurrence of severe outbreak every year [3]. The average yield loss caused by leaf blight (spot blotch, tan spot and *Alternaria* blight) in South Asia is around 20% but yield losses between 20% and 80% have been reported [4]. Under severe conditions, the yield losses may be as high as 100% [5]. Dormant conidia of *B. sorokiniana* respond poorly to fungicides and hence, seed treatment is not very effective in eliminating the pathogen which necessitates the identification of resistant genotypes as the most viable strategy. The resistance to spot blotch in emmer wheat is not well documented and is non-free threshable due to fragile rachis. Hence, a set of dicoccum lines were crossed to produce 18 F_1s which were subjected to mutagenesis to isolate free threshable dicoccum lines with resistance to spot blotch and isolated 55 free threshing dicoccum mutants [6].

The experimental material consisted of 55 advanced mutant lines along with 5 parents (MACS-2925, MACS-2912, MACS-2336, MACS-2931, DDK-1013 and DDK-1001) and 4 checks (DWR-162 (*T. aestivum*), DWR-1006 (*T. durum*), DDK-1001 (*T. dicoccum*) and NP-200 (*T. dicoccum*) which were sown in the field during *rabi* 2005-06 and 2006-07 to identify the spot blotch resistant dicoccum lines under natural epiphytotic conditions available at ARS, Arabhavi. Each line was sown in three rows measuring 6 m length with 23 cm between rows in an 8 x 8 simple lattice square design with two replications following the procedure of Cochran and Cox [7]. The lines were screened against the spot blotch by following double-digit system [8]. The reducing and non-reducing sugars were estimated in resistant and susceptible genotypes following Nelson's modification of Somogyi's method [9]. Estimation of total phenols present in plant samples was done following Folin-Ciocalteau reagent method [10]. Protein estimation was done by the procedure of Lowry *et al*. [11]. Bovine serum album was used as the standard. Morpho-physiological characters like stomatal frequency on upper surface (SUS) and stomatal frequency on lower surface (SLS) of different genotypes were studied at 40 and 80 days after sowing (DAS) [12].

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The screening of 64 genotypes including 55 free threshable dicoccum mutants for spot blotch resistance at ARS, Arabhavi under natural epiphytotic conditions resulted in the identification of 4 resistant genotypes namely NP-200 x DDK 1009-1, NP-200 x DDK-1009-9, NP-200 x MACS-2912-1 and DDK-1013x DDK-1001-3 (Table 1). Of the remaining genotypes 17 showed moderately resistant (MR) and moderately susceptible (MS) reactions, 25 showed susceptible (S) reactions and the check variety DDK-1009 showed highly susceptible (HS) reaction. Four free threshable lines that were resistant showed increased level of total sugars, reducing sugars, non-reducing sugars, phenol and protein and less number of stomatal frequencies (Table 2). The high phenol content in resistant genotypes may be due to more sugar as it acts as precursor for synthesis of phenols. Increased concentration of total phenols in callus tissues of sorghum genotypes has been reported to be responsible for resistance against downy mildew [13]. The phenol content in different genotypes

Table 1. Mean performance of advanced breeding lines of dicoccum wheat for leaf blight resistance over two years

Genotypes	LB (%)	Genotypes	LB (%)	Genotypes	LB (%)
Genotypes showing MR reactions		**Genotypes showing MS reactions**		**Genotypes showing S reactions**	
NP-200 x DDK-1001 -1	38	NP-200 x DDK-1001 -4	60	NP-200 x DDK-1009 -12	86
NP-200 x DDK-1001 -2	46	NP-200 x DDK-1009 -7	60	NP-200 x DDK-1009 -13	81
NP-200 x DDK-1001 -3	45	NP-200 x DDK-1009 -8	56	NP-200 x DDK-1009 -14	84
NP-200 x DDK-1009 -2	42	NP-200 x DDK-1009 -9	58	NP-200 x DDK-1009 -15	83
NP-200 x DDK-1009 -3	39	NP-200 x DDK-1009 -10	65	NP-200 x DDK-1009 -16	85
NP-200 x DDK-1009 -4	35	NP-200 x DDK-1009 -11	60	NP-200 x DDK-1009 -17	89
NP-200 x DDK- 1009 -5	37	NP-200 x MACS-2336 -1	62	NP-200 x DDK-1009 -18	83
NP-200 x DDK-1009 -6	38	DDK-1013 x DDK-1001 -4	60	NP-200 x DDK-1009 -19	78
NP-200 x MACS-2912 -2	40	DDK-1001 x MACS-2928 -2	64	NP-200 x DDK-1009 -20	88
NP-200 x MACS-2912 -3	39	DDK-1009 x MACS-2931 -3	65	NP-200 x DDK-1009 -21	85
DDK-1013 x DDK-1001 -1	38	DDK-1009 x MACS-2931 -4	63	NP-200 x DDK-1009 -22	80
DDK-1013 x DDK-1001 -2	46	DDK-1009 x MACS-2931 -5	59	NP-200 x DDK-1009 -23	84
DDK-1001 x MACS-2928 -1	45	Parents		NP-200 x MACS-2912 -4	81
DDK-1009 x MACS-2931 -1	40	MACS-2912	57	NP-200 x MACS-2912 -5	86
DDK-1009 x MACS-2931 -2	44	MACS-2336	68	NP-200 x MACS-2912 -6	87
Parent		MACS-2931	64	NP-200 x MACS-2912 -7	87
DDK-1013	38	Checks		NP-200 x MACS-2912 -8	85
Check		DWR-162 (*T. aestivum)*	63	NP-200 x MACS-2912 -9	89
NP-200 *(T. dicoccum)*	40	DWR-1006 *(T. durum)*	60	NP-200 x MACS-2336 -2	82
				NP-200 x MACS-2336 -3	83
Genotypes showing R reactions		**Highly susceptible (HS) genotype**			
NP-200 x DDK-1009 -1	12	DDK-1001 *(T. dicoccum)*	99	DDK-1009 x MACS-2931 -6	88
NP-200 x DDK-1009 -9	10			DDK-1009 x MACS-2931 -7	87
NP-200 x MACS-2912 -1	15			DDK-1009 x MACS-2931 -8	85
DDK-1013 x DDK-1001 -3	12			DDK-1009 x MACS-2931 -9	83
				Parent	
				MACS-2925	80
				Mean	62.4
				C.V. (%)	4.53
				C.D. (0.05)	5.53

Table 2. Mean values of the biochemical and morpho-physiological characters for leaf blight resistant lines at 40 and 80 DAS

Genotypes	Biochemical and morpho-physiological characters												
	40 DAS							80 DAS					
	TS	RS	NRS	Ph	Pr	SUS	SLS	TS	RS	NRS	Ph	Pr	SUS	SLS
NP-200 x DDK-1009-1 (5)	15.4	12.3	2.8	2.5	7.7	124.0	147.0	18.3	13.8	4.1	3.0	8.7	124.2	139.3
NP-200 x MACS-2912-1 (29)	15.3	12.2	2.6	2.7	7.3	128.5	146.9	18.2	13.6	4.5	3.4	8.4	124.5	139.9
NP-200 x DDK-1009-9 (14)	16.5	13.3	2.7	2.5	7.7	122.1	142.0	17.7	13.5	3.8	3.4	8.2	123.5	139.1
NP-200 x MACS-2912-3 (43)	16.5	13.3	2.7	2.5	7.7	122.7	125.9	19.0	14.8	4.6	3.6	8.5	122.0	123.5
Highly susceptible check														
DDK-1001 (*T. dicoccum*)	14.7	11.5	2.3	2.1	6.9	133.3	154.5	17.5	12.9	3.9	2.5	7.9	134.0	148.9
Mean	15.5	12.4	2.6	2.4	7.4	127.3	145.1	18.0	13.6	4.1	3.1	8.3	127.0	142.4
C.V. (%)	2.15	2.68	3.44	4.17	2.18	1.65	2.98	1.33	2.11	3.34	6.39	1.61	1.76	1.44
C.D. (0.05)	0.86	0.85	0.23	0.26	0.41	5.40	11.13	0.61	0.74	0.36	0.50	0.34	5.74	5.28

TS: Total sugar, RS: Reducing sugar, NRS: Non-reducing sugar, Ph: Phenol, Pr: Protein, SUS: Stomatal frequency on upper surface, SLS: Stomatal frequency on lower surface

significantly increased with increase in age of the plant. However, the amount of phenol content was significantly more in resistant genotypes compared to susceptible ones. Similarly, Harmas and Treba [14] reported high phenolic content in resistant barley and wheat genotypes against brown rust. The protein biosynthesis of the host has been widely assumed to be significant feature of pathogenesis, particularly during incompatible reaction. Quantitatively the total protein synthesis is much enhanced in the healthy tissues around the infected tissues. This additional protein is considered to be entirely of host origin. In the present findings mean soluble protein content was more in resistant genotypes of all three groups of wheat. Protein content increased slightly from 40 to 80 DAS. The resistant genotypes recorded more of soluble protein than the susceptible ones. Variation in soluble protein content at 40 DAS may be due to inherent character of these genotypes.

The stomatal frequency was higher in susceptible genotypes. More number of stomata was recorded on abaxial surface than that of adaxial surface of leaf, which indicates that, these act as main avenue for the entry of the pathogen into leaf tissue. Susceptible genotypes recorded higher frequency that provides higher opportunity for penetration by pathogen and results in high disease severity than resistant ones, where they recorded significantly lesser frequency and size of stomata [15]. Thus it appears that the number and size of the stomata are important characters of the leaf in relation to resistance or susceptibility of the plant to many foliar pathogens. Mutagen treatment followed by hybridization not only released wide variability but also resulted in novel variants with free threshability, resistance to spot blotch and essential quality parameters of emmer wheat. The four resistant lines NP-200 x DDK 1009-1, NP-200 x DDK-1009-9, NP-200 x MACS-2912-1 and DDK-1013x DDK-1001-3 could be further tested for their yield performance over locations for their utility as commercial varieties. Alternatively, they can be used in breeding programme to develop high yielding spot blotch resistant emmer wheat varieties for the popularization of its cultivation in nontraditional areas also due to its nutritional superiority than other forms of wheat.

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