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PHENOTYPING OF F_2 SEGREGATING POPULATION FOR POWDERY MILDEW TOLERANCE IN LINSEED (*LINUM USITATISIMUM* L.)

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Abstract

The biological experimental materials comprised of five powdery mildew resistant genotypes RLC-92, Sagar Local, Sabour Yellow, Sakoor and Neela were crossed as per Line × Tester design with four powdery mildew susceptible T-397, Chambal, Neelam and Shekhar genotypes, which was taken from AICRP on Linseed, Department of Genetics and Plant Breeding, I.G.K.V., Raipur (C.G.), India; during *Rabi* 2014-15 and 2015-16. In India and Chhattisgarh, it has been observed that major limiting factors for higher production is powdery mildew. Particularly in Chhattisgarh, the yield losses due to powdery mildew may be more than 60% when the disease is severe. Powdery mildew is the major cause in the linseed production during *utera* cultivation, It lows yield about 40%. The assessment of the disease per plant was obtained by observing the intensity of lesions present on the leaves. Keeping in this view, disease screening studies were made to understand the development of powdery mildew diseases. So, we need a high yielding linseed variety for late sown conditions with resistance to powdery mildew. With this objective, field screening of parents and twenty crosses for resistance to powdery mildew was initiated. Powdery mildew score ranged from 0 (free) to 5 (highly susceptible), 2 genotypes found highly resistant, 3 genotypes showed resistant, 3 genotypes comes under moderately resistant, 3 genotypes shows moderately susceptible, 5 genotypes showed susceptible and 13 genotypes shows highly susceptible. Highly resistant genotype could be utilized as donar parent for powdery mildew resistance breeding programme.

Key words : Linseed, powdery mildew, line × tester, resistance, susceptible, ALA, SDG, lignin.

Introduction

Linseed (*Linum usitatissimum* L.) 2n = 30, is an important oilseed crop that belongs to the genus *Linum* of the family Linaceae. It is also called flax or flaxseed. The name *Linum* originated from *lin* or "thread" and the species name *usitatissimum* is a Latin word meaning "most useful". It has been grown from ancient times for fiber (flax) and for its seed which is rich in oil. On the basis of diversity of plant types, linseed has two centers of origin *i.e.,* South West Asia, particularly in India (Vavilov, 1935; Richharia, 1962) and the Mediterranean region of Europe.

Around the globe linseed crop occupies an area of 22.70 lakh ha yielding out 22.39 lakh tones having an average productivity of 986 kg/ha. In India, it is grown in an area of 29210 ha with production and productivity being 141200 tones and 484 kg/ ha respectively. India ranks second in area after Canada, which is almost equivalent to China, which so far occupied the second slot in world area by the crop. Our national production slides to third place after Canada and China. India contributes about 14.89% and 6.56% to world area and production, respectively. In India, the crop is mainly cultivated in the states like Madhya Pradesh, Chhattisgarh, Uttar Pradesh, Maharashtra, Bihar, Odisha, Jharkhand, Karnataka and Assam accounting for more than 97 per cent of the total area. Chhattisgarh is one of the important linseed growing states of India, which account 26200 hectares area and 1100 tonnes production with productivity of 424 kg/ha (Annual Report, Linseed 2014-15).

Industrial oil and mostly, 80 percent of oil is used for paints, varnishes, a wide range of coating oils, linoleum, pad and printing inks, oil cloth, patent leather, enamels, stickers, tarpaulins leather and soap industries. Linseed contains about 33 to 45% oil and 24% crude protein which is one of the oldest commercial oils used for various purposes. Recently, it has gained a new interest in the

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emerging market of functional food due to its high content of fatty acids, alpha linolenic acid (ALA), an essential Omega3 fatty acid and lignan content or SDG (seco isolariciresinol diglucoside) which constitute about 57 % of total fatty acids in linseed (Morris, 2005). Almost every part of its plant is commercially utilized, either directly or after processing. New industrial uses of both the linseed oil and the fibres of oilseed flax also will increase the demand for this multi-use oilseed crop.

The crop is affected by some diseases like alternaria blight, powdery mildew, rust and wilt. powdery mildew, caused by the obligate biotrophic ascomycete *Oidium lini* Skoric, is a common, widespread and easily recognized foliar disease of flax present in most growing areas worldwide (Gill, 1987; Beale, 1991; Saharan and Saharan, 1998; Aly *et al*., 2012) is one of the major limiting factors of linseed. The disease appear on all the aerial parts of the plant, ultimately causes substantial losses in yield up to 60% (Srivastava *et al*., 1997; Reddy *et al*., 2013a). Over the last two decades, the importance of this disease has increased probably due to the appearance and rapid distribution of new races capable of attacking the previously resistant cultivars (Aly *et al*., 1994; Mohamed, 2012). In India and Chhattisgarh, it has been observed that major limiting factors for higher production is powdery mildew. Particularly in Chhattisgarh the yield losses due to powdery mildew may be more than 60% when the disease is severe, particularly at the time of flower initiation.

Despite considerable increase in productivity and production a wide gap exists between potential yield and the yield realized at farmer's field, which is largely because of a number of biotic stresses, to which linseed crop is exposed. The cost of chemicals farmer rarely practice such control measures and the usage of such fungicide will negatively affect environment and specially human health. Therefore, the most effective way to control powdery mildew is the use of resistant varieties. Genetic resistance is a priority for flax breeders because fungicides can be hazardous, costly, and associated with environmental concerns. The genetics of resistance to powdery mildew in flax and reported a single dominant gene designated PM1 for resistance to powdery mildew (Rashid and Duguid, 2003).

Therefore, there is a need to develop varieties resistant to powdery mildew to stabilize the yield potentials of linseed varieties. Therefore, this research work helps in developing varieties resistant powdery mildew diseases to stabilize the yield potentials of linseed varieties. The manipulation of inherent potentials of plants in the form of resistant varieties is a cheap, viable and environment friendly alternative to reduce losses from biotic stress.

Materials and Methods

The biological experimental materials comprised of five powdery mildew resistant genotypes RLC-92, Sagar Local, Sabour Yellow, Sakoor and Neela were crossed with four powdery mildew susceptible T-397, Chambal, Neelam and Shekhar genotypes, which was taken from AICRP on Linseed, Department of Genetics and Plant Breeding IGKV, Raipur (C.G.), India. The parentage of genotypes is presented in table 1. The crosses were attempted as per Line × Tester design during *Rabi* season 2014-2015. An F_1 population from a cross between powdery mildew resistance and powdery mildew susceptible was generated, for advancement of F_1 population, seeds were grown in pots in a greenhouse and sufficient F_2 seeds were produced. In the next *Rabi* season 2015-2016 parents, F_1 and F_2 was grown and genetic studies was done for powdery mildew. All recommended package of practices was followed except spraying of plant protection chemical to allow maximum inoculation of powdery mildew diseases.

Disease assessment

Data on disease severity of powdery mildew on leaves was recorded on 10 plants sampled randomly from each cross and disease scoring was done as per the scale of AICRP (1991) (table 2).

Screening method

Parents and crosses was screened against powdery mildew under natural epiphytotic conditions during *Rabi* 2015-16 under the two environments *viz.* Block- 1 and Block 2 at the Research cum Instructional Farm, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh, India. The intensity of disease in the field was estimated from ten randomly selected plants in each crosses which were tagged with labels, Each plant was scored visually in the field and plants were rated in 0-5 scale, where, 0- highly resistance, 1- resistance, 2- moderately resistance, 3- moderately susceptible, 4- susceptible and 5-highly susceptible (AICRP, 1991). The assessment of the disease per plant was obtained by observing the intensity of lesions present on the leaves. The plants with disease rating <2 were considered as resistant and above 2 as susceptible. All recommended package of practices was followed except spraying of plant protection chemical to allow maximum inoculation of powdery mildew diseases. The observations were recorded based on disease reaction, plants of each cross were classified as breeding true for resistance, true for susceptible or segregating for resistant and susceptible.

Table 1 : Characteristic features of parents.

S. no.	Line/ Tester	Origin	Parentage	Special features
	Lines			
1	RLC-92	IGKV,Raipur	Jeevan× LCK 9209	Resistant to powdery mildew, tall, violet flower, brown seeded, oil content- 39%.
2	Sagar Local	Sagar,M.P.	Local selection	Resistant to powdery mildew, Red violet flower
3	Sabour Yellow	RAU,Bihar	Local selection	Resistant to powdery mildew, Red violet flower
4	Sakoor	GMUKanpur	Local selection	Resistant to powdery mildew, violet flower
5	Neela	W.B.	Local selection of W.B.	Resistant to powdery mildew, Brown, medium seeded, Oil content - 40%.
	Testers			
1	T-397	Kanpur	T491 x T1193-1	Susceptible to powdery mildew, brown seeded, Oil content - 44%.
2	Chambal	Rajasthan	Local x RR45	Susceptible to powdery mildew, brown, large seeded, Oil content - 44%.
3	Neelam	Kanpur	T1 x NP(RR)9	Susceptible to powdery mildew, brown seeded, Oil content - 43%.
4	Shekher	Kanpur	Laxmi 27 x EC1387	Susceptible to powdery mildew, Shiny brown seed, Oil content - 43%.

Table 2 : Scale (0-5) for rating of reaction to powdery mildew.

S. no.	Score	Disease intensity (% area of leaves/ plant infected)	Rating	
1	0	Free from disease	Highly resistant	HR
2	1	1 to 10	Resistant	R
3	2	11 to 25	Moderately resistant	MR
4	3	26 to 50	Moderately susceptible	MS
5	4	51 to 75	Susceptible	S
6	5	Above 75	Highly susceptible	HS

Results and Discussion

The parents and twenty crosses were screened against powdery mildew under natural epiphytotic conditions during *Rabi* 2015-16 under the two environments *viz.* Block no.1 and Block no.2, Experimental Farm of College of Agriculture, Raipur (C.G.), India. Parents, F_1 and F_2 of twenty crosses were sown in second week of november. The intensity of disease in the field was estimated from ten randomly selected plants in each crosses which were tagged with labels. Each plant was scored visually in the field and plants were rated in 0-5 scale where, 0- highly resistance, 1- resistance, 2- moderately resistance, 3- moderately susceptible, 4- susceptible and 5-highly susceptible (AICRP, 1991). The assessment of the disease per plant was obtained by observing the intensity of lesions present on the leaves. The plants with disease rating <2 were considered as resistant and above 2 as susceptible. The observations were recorded based on disease reaction, plants of each cross were classified as breeding true for resistance, true for susceptible or segregating for resistant and susceptible. All recommended package of practices was followed except spraying of plant protection chemical to allow maximum inoculation of powdery mildew diseases. The powdery mildew disease incidence was quite severe during the season due to conditions favourable for the development of the disease. Parent and crosses were screened for their reaction against powdery mildew diseases and depending upon their

Table 3 : Reaction of parents of linseed to powdery mildew (*Oidium lini*) over two environments under natural epiphytotic conditions during 2015-16 Raipur, C.G.

S. no.	Genotypes	Reaction	Disease score at particular environment	
			Block 1	Block 2
1	RLC-92	Resistant	0	0
2	Sagar Local	Resistant	0	1
3	Sabour Yellow	Resistant	0	1
4	Sakoor	Resistant	0	0
5	Neela	Resistant	1	2
6	T-397	Susceptible	5	4
7	Chambal	Susceptible	5	5
8	Neelam	Susceptible	5	5
9	Shekhar	Susceptible	4	5

Table 4 : Reaction of the F_2 crosses to powdery mildew(*Oidium lini*) over two environments under epiphytotic conditions during 2015-16 Raipur, C.G.

S. no.	F_2 segregating generations	Disease score at particular environment	
		Block no 1	Block no 2
1	RLC-92 xT-397	1	2
2	RLC-92 x Chambal	3	4
3	RLC-92 x Neelam	4	5
4	RLC-92 x Shekhar	0	4
5	Sagar Local x T-397	2	3
6	Sagar Local x Chambal	4	5
7	SagarLocal x Neelam	4	5
8	SagarLocal x Shekhar	2	3
9	Sabour Yellow x T-397	3	5
10	Sabour Yellow x Chambal	3	5
11	Sabour Yellow x Neelam	4	5
12	Sabour Yellow x Shekhar	4	3
13	Sakoor x T-397	3	5
14	Sakoor x Chambal	4	4
15	Sakoor x Neelam	4	5
16	Sakoor x Shekhar	2	1
17	Neela x T-397	2	2
18	Neela x Chambal	3	4
19	Neela x Neelam	5	5
20	Neela x Shekhar	4	5

genetic makeup of each crosses responded differentially to powdery mildew diseases. Environmental factors such as humidity, temperature or light can influence development of the diseases possibly explaining these differences in scores. The results are based on screening under natural field conditions so, the resistant genotypes need to be evaluated under artificial conditions to confirm the resistance before using them in breeding programme. Despite being high susceptible, some crosses produced good yield and showed tolerance to powdery mildew disease. Highly resistant genotype could be utilized as donar parent for powdery mildew resistance breeding programme.

Powdery mildew appeared at both the locations during *rabi* 2015-16.Data on reaction to powdery mildew under epiphytotic conditions for nine parents and twenty F_2 crosses over the two locations *viz.* Block 1 and Block 2 under study is presented in tables 2, 3 and 4.

From the results (rable 5), it is concluded that two parents *viz*. RLC-92 and Sakoor (Disease score 0 at all the locations) were highly resistant to the disease, one parent and two F_2 crosses *viz*. Sagar Local, SabourYellow, Sakoor × Shekhar (Disease score 1 or <1) were found to be resistant, one parents and two F_2 crosses namely Neela, RLC-92 × T-397, Neela × T-397 (disease score 2 or <2) being moderately resistant, three F_2 crosses *viz*. Sagar Local × T-397, SagarLocal × Shekhar, Sabour Yellow × Shekhar (disease score 3 or <3) were moderately susceptible, one parent and four crosses *viz.* T-397, RLC-92 × Chambal, RLC-92 × Shekhar, Sakoor × Chambal, Neela × Chambal (disease score 4 or <4) were found to be susceptible and three parents and ten F_2 crosses *i.e.* Chambal, Neelam, Shekhar, RLC-92 × Neelam, Sagar Local × Chambal, Sagar Local × Neelam, Sabour Yellow × T-397, Sabour Yellow × Chambal, Sabour Yellow × Neelam, Sakoor × T-397, Sakoor × Neelam, Neela × Neelam, Neela × Shekhar (disease score 5 or <5) were found to be highly susceptible to powdery mildew.

The genotype categorized as highly resistance or resistant to powdery mildew need further evaluation under artificial condition for their use as source of

Table 5 : Reaction of the parents and crosses of linseed to powdery mildew*(Oidium lini*) during 2015-16 Raipur, C.G., India.

S. no.	Disease score	Reaction	No. of genotype	Genotypes
1	0	Highly resistant(HR)	2	RLC-92,Sakur
2	1	Resistant(R)	3	Sagar Local, Sabour Yellow, Sakoor x Shekhar
3	2	Moderetely resistant(MR)	3	Neela, RLC-92 xT-397, Neela x T-397
4	3	Moderetely susceptible(MS)	3	Sagar Local × T-397,Sagar Local × Shekhar, Sabour Yellow × Shekhar
5	4	Susceptible(S)	5	T-397, RLC-92 × Chambal,RLC-92 × Shekhar, Sakoor × Chambal, Neela × Chambal
6	5	Highly susceptible(HS)	13	Chambal, Neelam, Shekhar, RLC-92 × Neelam, Sagar Local × Chambal, Sagar Local × Neelam, Sabour Yellow × T-397, Sabour Yellow × Chambal, Sabour Yellow × Neelam, Sakoor × T-397, Sakoor × Neelam, Neela × Neelam, Neela × Shekhar

resistance in the future breeding programmes. These results are in agreement with the findings of Pathania (1999), who found three genotypes of linseed (Jeewan, Kiran and Flak-1) as resistant to powdery mildew. Sharma *et al*. (1972) reported that none of the 104 varieties evaluated were resistant against *Oidium lini* but 45 were considered moderately resistant and remaining varieties were moderately susceptible. Gupta and Jain (1980) screened the material for two years under natural conditions and found EC-9832 genotype was highly resistant and M-10 was moderately resistant. Basandrai *et al*. (1994) evaluated indigenous and exotic genotypes of flax for resistance to rust (*Melampsora lini*), wilt (*Fusarium oxysporum*) and powdery mildew (*Oidium lini*) and reported 139,35 and 24 genotypes, respectively, being free of infection. Twenty two genotypes were free from or resistant to all three diseases. Rashid and Duguid (2005) investigated on the genetics of resistance to powdery mildew in flax and identified several resistant Canadian and introduced cultivars. Kumar (2015) found that 16 genotypes *viz*. Jeewan, Nagarkot, Ariane, Hearmies, Janaki, Him Alsi-2, Binwa, Baner, Mariena, Giza-6, Giza-7, Giza-8, Faking, Ayogi, Flak-1 and KL-241 were resistant to powdery mildew as well as rust. Similar result also reported by Prasad *et al*. (1988), Ashry *et al*. (2002), Mohamed *et al*. (2012) and Dash *et al*. (2016).

Conclusion

Plant breeders need to explore the nature of disease resistance and identify additional resistance genes from new sources. Therefore, this research work helps in developing varieties resistant to powdery mildew to stabilize the yield potentials of linseed varieties.

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