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Breeding Strategies for Simultaneous Improvement in Anthracnose Disease Resistance and Economically Important Traits in French Bean

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ABSTRACT

Background: French bean (*Phaseolus vulgaris* L.) production in the tropics is threatened by heavy incidence of anthracnose disease causing substantial crop loss when infected at the early growth stages. Breeding strategies for enhanced productivity, high pod protein content combining resistance to anthracnose disease needs attention and are to be formulated.

Methods: Diverse genotypes with broad genetic base and phenotypic diversity in five bush and one pole types were crossed in a 6 × 6 half diallel mating design to estimate combining ability, mode of gene action and extent of heterosis for 11 quantitative traits. Statistical analyses were done with Windostat (ver. 8.0, Indostat Services, Hyderabad, India).

Result: The additive genetic effect was evident for percent disease index (PDI) of anthracnose and ten pod weight. Rapid genetic gain can be achieved due to predominance of additive gene action and can therefore be selected in early generation through simple breeding methods. The remaining economic traits controlled by additive and non-additive gene effects could be improved through biparental mating, reciprocal recurrent selection, or diallel selective mating. Anthracnose disease resistant and high yielding cultivars can be developed utilizing good general combiners *viz*., Laxmi, Arka Sharath and Vaishnavi-264 with high gca effects. Although two cross combinations Arka Sharath × Lakshmi and Arjun × Arka Sharath showed significant heterobeltiosis in desired direction for PDI of anthracnose and other desirable horticultural traits but could not be exploited at commercial level due to complexity in hybridization. Identifying pure lines with resistance against anthracnose disease and favourable horticultural attributes could be accomplished in segregating generations of the prospective hybrids.

Key words: Anthracnose disease, Combining ability, Gene action, *Phaseolus vulgaris*.

INTRODUCTION

In terms of morphological polymorphism, applications and the range of habitats to which they have been adapted, French bean (*Phaseolus vulgaris* L.) is an incredibly diversified crop (Basavaraja *et al*., 2021), often considered as the "poor man's meat" (Kargiotidou *et al*., 2019). The current cultivars of French bean have lower productivity, non-synchronous flowering and fruiting, lodging and shattering susceptibility and complete or partial absence of genetic resistance to significant insect pests and diseases that caused significant damage and very poor harvest indexes (Immaculee, 2011). Due to complicated flower structure and issues with poor cross pod setting, hybrids are not commercially exploited. The crop's productivity is consequently far lower in India (9.96 mt/ha) than the global average (16 mt/ha) (Anonymous, 2021; FAOSTAT, 2017). One of the main reasons of low productivity is high incidence of anthracnose disease [*Colletotrichum lindemuthianum* (Sacc. and Magnus) Briosi and Cavara] which causes poor seed germination and seedling vigor and more plant death. Yield losses can exceed 100% when badly contaminated seed is sowed in cold, rainy weather, which is conducive to the development of disease (Sharma *et al*., 1994). This is because the fungus overwinters inside bean seeds. Planting resistant cultivars is the most efficient, cost-efficient and farmer-friendly method of managing this disease (Maibam *et al*., 2015; Prabha *et al*., 2020; Ganie and Wani, 2022).

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Evaluation of available French bean germplasm to identify acceptable and resistant genotypes is necessary to improve its yield. In a self-pollinated crop, an objective evaluation of a particular cross would primarily depend on its hybrid vigour and combining ability (Fasoulas, 1980), as well as on accurate estimates of various genetic components such as additive, dominance, non-allelic interactions, linkage among the polygenes and gene dispersion in the parents of a cross (Jinks, 1983). To achieve this goal, diallel (Griffing, 1956) is one such analysis which is a useful tool for preliminary evaluation of genetic stock for use in hybridization programme with a view to identify good general as well as specific combiners. Such studies also simultaneously illustrate the nature and magnitude of gene action involved in the expression of desirable traits. The experiment was conducted to identify the best combiners and to determine the gene action for controlling anthracnose disease severity and economically important traits in French bean to formulate a breeding strategy.

MATERIALS AND METHODS

The investigation was carried out at Research Farm of Bidhan Chandra Krishi Viswavidyalaya, Kalyani, Nadia, West Bengal, India, situated at 23°N latitude and 89°E longitude at a mean sea level of 9.75 m.

Seeds of six parental genotypes (‘Vaishnavi-264’, ‘Arjun’, ‘Falguni’, ‘Poorva’, ‘Arka Sharath’, bush types and ‘Lakshmi’, pole type), after treating with thiram @ 3 $g.kg^{-1}$ of seed, were sown in well-prepared flat beds at a spacing 45 × 30 cm on 25th October, 2020. Well decomposed FYM @ 10 $ton.ha^{-1}$ and N:P:K @ 100:60:50 $kg.ha^{-1}$ was applied during land preparation. During the full bloom, the parental lines were crossed in a half diallel fashion. The crossed pods were harvested when dried. Cleaned F_1 seeds were kept in desiccator for next season sowing.

Seeds of 15 F_1’s and 6 parental lines were sown in well-prepared beds following the previous methods in a randomized block design with three replications during 24 October, 2021. Normal package of practices were followed to raise a good crop (Chattopadhyay *et al*., 2007).

The severity of bean anthracnose was recorded in each parental line and F_1 starting from 30 days after sowing (DAS) up to 90 DAS following the disease rating scale (0-9) adopted by Mayee and Dattar (1986) and Percent Disease Index (PDI) was calculated from the numerical ratings by the formula suggested by McKinney and Davis (1925).

Observations were recorded on days to 50 % flowering, pod length (cm), pod diameter (cm), number of pods per plant, ten pod weight (g), 100 seed weight (g), number of seeds per pod, green pod yield per plant (g) from 15 randomly selected plants from each plot for each replication. Total soluble pod protein was estimated as per of Lowry *et al*. (1951). Total sugar content of green pod was estimated following anthrone method as per DuBois *et al*. (1956).

Data were analyzed following Gomez and Gomez (1984). The magnitude of heterosis was estimated in relation to better-parent (BP) values according to Hayes *et al*. (1965). Combining ability variances and effects, were determined according to Griffing (1956) Model 1 and Method 2. Dominance estimates (D.E.) were computed using the formula of Smith (1952). Statistical analyses were done with Windostat (ver. 8.0, Indostat Services, Hyderabad, India).

RESULTS AND DISCUSSION

Gene action of quantitative traits

The analysis of variance for combining ability based on Griffing’s Model 1 and Method 2 exhibited significant components of GCA and SCA mean squares for PDI of anthracnose and other economically important traits in F_1 generation (Table 1) suggesting apparent control of both additive and non-additive gene action.

The relative importance of additive and non-additive genetic effects for a trait is reflected by the predictability ratio (Baker, 1978). Predictability ratio was >0.80 for PDI of anthracnose and ten pod weight indicating the role of additive gene action in control of these traits (Table 1). Recurrent selection may increase the frequency of favorable alleles and identify superior cross combinations by repeated crossing and selection in later generations which would be the best possible approach to exploit additive gene effects for the improvement of traits. The economically important traits *viz.* days to 50 % flowering, pod length, pod diameter, number of pods per plant, 100 seed weight, number of seeds per pod, green pod yield per plant, total soluble pod protein content and total sugar content of green pod, were controlled both by additive and non-additive gene action as their predictability ratios were ≥0.50 and <0.80 (Table 1). A population improvement approach in the form of diallel selective mating (Jensen 1970) or mass selection with concurrent random mating (Redden and Jensen, 1974) or restricted recurrent selection by intermating the most desirable segregates followed by selection (Shende *et al*., 2012) could be followed for the exploitation of additive and non-additive gene action governing traits.

Combining ability analysis

Among the parents none was found to be a good general combiner for all the characters under study (Table 2). Among the parents, the maximum significant GCA effects in desired directions were recorded by the genitors, Arka Sharath, Vaishnavi and Lakshmi for PDI of anthracnose and other economically important traits including pod yield per plant. The minimum disease severity of anthracnose and the maximum average performance for pod yield per plant along with other economic traits was recorded in Arka Sharath followed by Lakshmi and Vaishnavi (Table 2). These three genitors could be identified as potential donors because they exhibited the highest frequency of low severity of disease and high yielding cross combinations along with good quality attributes when crossed with other genitors.

Different cross combinations exhibited different SCA effects and only a few crosses expressed consistently either positive or negative SCA effects for certain traits (Table 3). High significant SCA effects in desired direction for PDI of anthracnose and other economic traits were shown by two crosses Arka Sharath × Lakshmi and Arjun × Arka Sharath which also involved either both parents or one of the parents as good general combiner(s) for PDI of anthracnose and other economic traits, suggesting further exploitation of these crosses in segregating generation to identify superior lines of fixable nature.

Study on heterosis

Heterobeltiosis and the mean performance for different characters varied (Table 3) among the crosses. The extent of heterobeltiosis was not so high in French bean in the present study. Maximum heterobeltiosis in desired direction with highest average performance for PDI of anthracnose and pod yield per plant was exhibited in the hybrid of Arka Sharath × Lakshmi and Arjun × Arka Sharath. Other horticultural traits exhibiting significant heterobeltiosis in the desired directions were protein content of pod (Arka Sharath × Lakshmi), total sugar (Vaishnavi-264 × Lakshmi), number of pods per plant (Arka Sharath × Lakshmi) and days to 50% flowering (Arjun × Arka Sharath). Although commercial exploitation of these promising hybrids could not be done as such but these hybrids could also be exploited in segregating generations to identify pure lines having appreciable resistance to anthracnose disease combining high yield. The manifestation of heterosis in some crosses was relatively high which might be due to the more diverse parents involved in these crosses or intermediate divergent than other parents that manifested little, or no, heterosis in their crosses. The absence of significant heterosis in desired direction in crosses with respect to pod diameter could be explained by internal elimination of heterotic components.

Based on gca effects, the promising heterotic crosses involved four types of combinations namely, H × H, H× L, L × H and L × L, where H denotes significant GCA effect of parent in desired direction and L stands for non-significant GCA effect of the parent (Table 4). In the H × H type cross combinations, additive as well as additive × additive type of interactions were involved. These crosses would be very useful as desirable segregates would be fixed in early advance generation. On the other hand, crosses of H × L type or L × H type involved at least one parent with significant GCA effect indicating predominantly the presence of additive genes in good combiner and possibly complementary epistatic effect in poor combiner and these two gene actions acted in complementary fashion to maximize the expression (Salimath and Bahl, 1985). In crosses involving L × L category, SCA effects seemed to have played a very important role and high performance was due to non-additive gene action (Bhutia *et al*., 2015).

Anthracnose disease severity

Disease severity of bean anthracnose is an essential criterion to judge the resistance level of French bean parents

Table 1: Analysis of variance (mean square) for combining ability (Griffing's model 1 and method 2).

Source of variation	df	Days to 50% flowering	Pod length (cm)	Pod diameter (cm)	Number pods per plant	10 pod weight (g)	100 seed weight (g)	Number seed per pod	Protein content (%)	Total sugar (%)	PDI[a] of bean anthracnose (%) at 90 DAS[b] (%)	Pod yield per plant (g)
GCA	5	45.318**	5.023**	0.111**	63.234**	853.96**	35.045**	5.6111**	1.503**	2.626**	8.53**	9179.18**
SCA	15	4.295**	0.502*	0.007**	10.426**	788.34**	3.493**	0.607**	0.206**	0.512**	0.443**	645.10**
Error	40	1.181	0.181	0.0002	1.314	53.187	0.591	0.161	0.003	0.0813	0.134	5.729
σ^2a[c]		11.034	1.210	0.027	15.479	213.159	8.613	1.362	0.375	0.636	2.098	2293.362
σ^2na		3.114	0.321	0.007	9.111	51.227	2.901	0.446	0.203	0.431	0.308	639.377
$\frac{\sigma^2a}{\sigma^2a+\sigma^2na}$		0.779	0.790	0.790	0.629	0.806	0.748	0.753	0.648	0.596	0.871	0.781

*, **Significant at $P \leq 0.05$ and $P \leq 0.01$ level, respectively.

[a]PDI= Percent disease index.

[b]DAS= Days after sowing.

[c]σ^2a= Additive genetic variance; σ^2na= Non-additive genetic variance; $\sigma^2a/(\sigma^2a+\sigma^2na)$ = Ratio of additive genetic variance to total genetic variance.

and hybrids. Reactions of parents and hybrids in terms of PDI values of bean anthracnose differed at different DAT (Fig 1 and Fig 2). All parents and hybrids showed comparatively lower PDI values from 30 to 60 DAS. An increase in PDI values occurred from 60 DAS up to 90 DAS. The PDI values were lower in Arka Sharath and Lakshmi among parents and Arka Sharath × Lakshmi, Arjun × Arka Sharath and Vaishnavi-264 × Arka Sharath in hybrids up to 120 DAT. Previous researchers have carried out screening and documented variable levels of anthracnose resistance in native land races and exotic French bean genotypes (Kour *et al*., 2012; Maibam *et al*., 2015; Ganie and Wani, 2022). Planting resistant cultivars is the most efficient, affordable and simple management method for this disease.

Dominance estimates

Dominance estimates values varied among the 15 F_1 crosses studied (Table 4). Different degrees of gene effects;

Table 2: Estimates of general combining ability (gi) effects in 6 parents over 15 F_1s with average performances.

Character	Vaishnavi-264	Arjun	Falguni	Poorva	Arka Sharath	Lakshmi	SE (si)
Days to 50% flowering	-0.362 (48.67)	0.598 (51.00)	1.723** (52.33)	3.223** (53.33)	-3.319** (42.67)	-1.862** (45.33)	1.397
Pod length (cm)	0.260 (15.57)	-0.073 (15.17)	-0.593** (14.24)	-1.156** (13.73)	1.062** (17.73)	0.501** (15.61)	0.547
Pod diameter (cm)	-0.023** (0.77)	-0.041** (0.77)	-0.074** (0.71)	-0.146** (0.643)	0.125** (1.107)	0.159** (1.09)	0.0189
Number pods per plant	0.363 (51.67)	-1.387* (49.67)	-2.554** (49.00)	-2.970** (48.67)	3.858* (56.33)	2.691** (54.33)	1.473
10 pod weight (g)	-0.636 (63.02)	-5.255** (53.28)	-8.256** (48.92)	-11.003** (47.30)	12.914** (86.20)	12.236** (80.29)	1.482
100 seed weight (g)	0.757** (27.05)	-0.689** (24.03)	-1.764** (23.35)	-2.773** (22.71)	2.057* (29.38)	2.413** (29.28)	0.988
Number seed per pod	0.167 (7.33)	-0.167 (6.33)	-0.708** (6.00)	-1.125** (5.67)	0.958** (8.67)	0.875** (8.33)	0.515
Protein content (%)	-0.012 (2.58)	0.057* (2.64)	0.544** (3.687)	0.394** (3.90)	-0.461** (1.81)	-0.522* (2.027)	0.066
Total sugar (%)	0.114 (6.33)	-0.264** (5.78)	-0.276** (5.71)	-0.818** (5.027)	0.764** (7.32)	0.479** (6.80)	0.366
PDI[a] of bean anthracnose at 90 DAS[b] (%)	-0.379** (13.76)	0.052 (14.87)	1.179** (16.71)	1.255** (16.73)	-1.060** (11.86)	-1.046** (12.34)	0.470
Pod yield per plant (g)	16.34**(321.02)	-1.543 (290.51)	-26.11** (254.75)	-52.46** (224.57)	31.30** (364.89)	32.47** (344.66)	3.076

*, **Significant at $P\leq0.05$ and $P\leq0.01$ level, respectively; Figures in parentheses indicate mean performance.

[a]PDI= Per cent disease index.

[b]DAS= Days after sowing.

Table 3: Promising cross combinations in F_1 generation based on gca and sca effects of French bean hybrids.

Character	Cross(es) with high heterobeltiosis in desired direction along with type of cross combination	Range of heterobeltiosis (%)	Significant heterobeltiosis (%)	Average performance with *sca* effect of the cross
Days to 50% flowering	Arjun × Arka Sharath (L × H)	-17.65 to -1.25	-17.65**	42.00 (-3.262**)
Pod length (cm)	Vaishnavi-264 × Lakshmi (L × H)	-16.79 to 9.67	9.67*	17.12 (0.452)
Number pods per plant	Arka Sharath × Lakshmi (H × H)	-4.73 to 20.63	20.63**	67.96 (7.156**)
	Vaishnavi-264 × Arka Sharath (L × H)		11.24**	62.67 (4.194**)
10 pod weight (g)	Vaishnavi-264 × Lakshmi (L × H)	-21.60 to 9.49	9.49**	87.92 (6.819**)
	Arka Sharath × Lakshmi (H × H)		9.34**	94.25 (-0.397)
100 seed weight (g)	Arka Sharath × Lakshmi (H × H)	-14.18 to	20.10**	35.29 (3.197**)
	Vaishnavi-264 × Arjun (H × L)		10.59*	29.91 (2.223**)
Number seed per pod	Arka Sharath × Lakshmi (H × H)	-23.08 to 19.23	19.23**	10.33 (0.786*)
Protein content (%)	Arka Sharath × Lakshmi (L × L)	-45.94 to 26.15	26.15**	2.56 (0.477**)
Total sugar (%)	Vaishnavi-264 × Lakshmi (L × H)	-10.87 to 25.82	25.82**	8.56 (1.119**)
	Arjun × Falguni (L × L)		19.54**	6.91 (0.606*)
	Vaishnavi-264 × Arka Sharath (L × H)		19.43**	8.75 (1.024**)
PDI[a] of bean anthracnose at 90 DAS[b] (%)	Arka Sharath × Lakshmi (H × H)	-16.18 to -0.04	-16.18**	9.94 (-1.145**)
	Arjun × Arka Sharath (L × H)		-13.74**	10.23 (-1.028**)
Pod yield per plant (g)	Arka Sharath × Lakshmi (H × H)	-36.70 to 10.45	10.45**	403.03 (24.875**)
	Arjun × Arka Sharath (L × H)		9.75**	400.50 (21.629**)

*, **Significant at $P\leq0.05$ and $P\leq0.01$ level, respectively. SCA = specific combining effect; H= Significant specific combining effect (GCA) effect of the parents in the desired direction; L= Negative/positive GCA effect of the parents in the opposite direction.

[a]PDI= Per cent disease index; [b]DAS= Days after sowing.

Table 4: Estimates of dominance effects of 11 characters of French bean.

Cross combination	Days to 50% flowering	Pod length (cm)	Pod diameter (cm)	Number pods per plant	10 pod weight (g)	100 seed weight (g)	Number seed per pod	Protein content (%)	Total sugar (%)	PDI[a] of bean anthracnose at 90 DAS[b] (%)	Pod yield per plant (g)
Vaishnavi-264 × Arjun	-3.29	7.85	17.50	5.00	0.01	2.89	2.33	11.43	2.65	-0.41	1.93
Vaishnavi-264 × Falguni	-0.27	0.47	0.43	0.75	0.33	0.76	1.00	1.22	0.06	-0.12	0.28
Vaishnavi-264 × Poorva	0.00	0.63	0.45	0.78	0.61	0.63	0.20	1.16	0.57	-0.26	0.36
Vaishnavi-264 × Arka Sharath	-1.11	1.02	1.20	3.72	1.41	2.47	2.50	1.03	3.85	-0.37	2.24
Vaishnavi-264 × Lakshmi	-0.41	84.72	1.19	4.00	1.88	2.93	2.33	1.73	8.43	-1.15	3.55
Arjun × Falguni	-1.00	0.94	-0.89	1.01	2.44	2.97	0.98	1.41	33.20	-0.38	0.42
Arjun × Poorva	-0.14	0.16	0.49	1.67	0.64	0.65	2.01	1.23	1.36	-1.99	0.18
Arjun × Arka Sharath	-1.16	0.44	0.66	1.40	1.00	1.57	1.86	1.73	1.09	-0.32	1.07
Arjun × Lakshmi	-0.18	6.22	1.13	1.86	1.33	1.45	2.00	1.33	1.88	-0.06	1.60
Falguni × Poorva	-1.01	0.62	0.27	1.00	0.34	0.37	2.98	3.29	1.90	0.30	0.43
Falguni × Arka Sharath	-0.59	0.65	0.61	0.91	0.58	0.72	0.50	0.85	2.14	-0.22	0.32
Falguni × Lakshmi	-0.52	2.35	0.95	1.37	1.05	1.09	1.00	0.18	1.49	-0.12	0.70
Poorva × Arka Sharath	0.88	-0.49	-0.63	0.31	0.04	-0.25	-0.33	-0.37	0.31	0.71	-0.91
Poorva × Lakshmi	-0.58	1.37	0.37	1.12	0.72	0.40	0.25	-0.91	1.01	-0.13	0.28
Arka Sharath × Lakshmi	-1.25	1.03	6.06	12.63	3.73	124.06	10.96	5.98	5.36	-0.67	4.77

[a]PDI= Per cent disease index.

[b]DAS= Days after sowing.

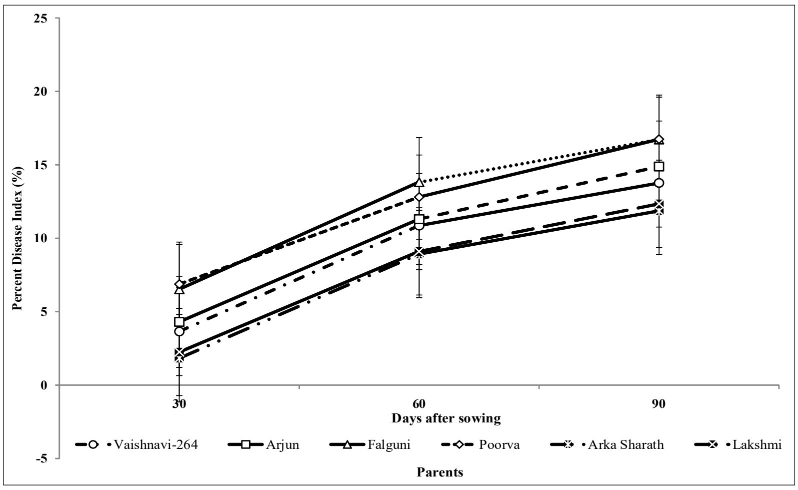

Fig 1: Per cent disease index (%) of anthracnose in parents from 30 to 90 days after sowing.

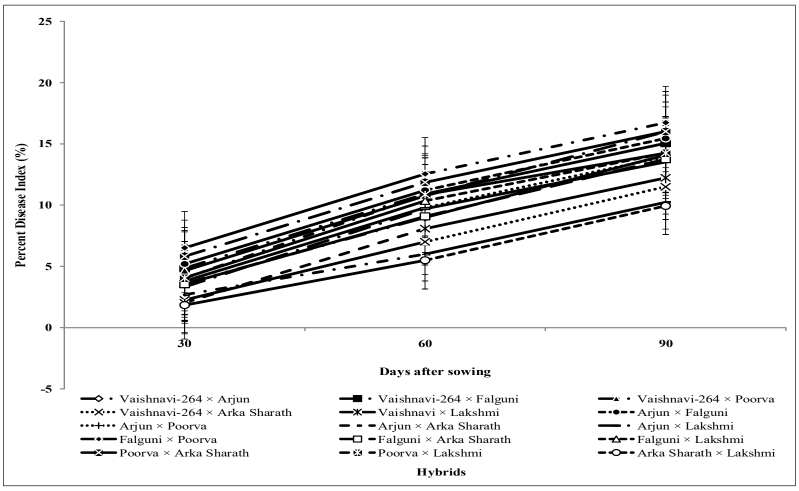

Fig 2: Per cent disease index (%) of anthracnose in hybrids from 30 to 90 days after sowing.

i.e., partial- to over-dominance, were involved in the inheritance of pod yield and its components, quality and anthracnose disease severity traits (Table 4). No previous studies have been documented so far in French bean to support the present findings.

CONCLUSION

Recurrent selection would be the best possible approach to exploit additive gene effects for the improvement of PDI of anthracnose and ten pod weight. While diallel selective mating or mass selection with concurrent random mating or restricted recurrent selection by inter-mating the most desirable segregates followed by selection could be followed for the exploitation of additive and non-additive gene action for rest of the economic traits. Three genitors, Arka Sharath, Lakshmi and Vaishnavi were found promising for future breeding. Two crosses Arka Sharath × Lakshmi and Arjun × Arka Sharath showed significant heterobeltiosis in desired direction for PDI of anthracnose, pod yield per plant and other desirable horticultural traits. Partial- to over-dominance, were found to be involved in the inheritance of anthracnose disease severity, pod yield and its components and quality traits. Although commercial exploitation of promising hybrids could not be done these hybrids could be exploited in segregating generations to identify pure lines having resistance to anthracnose disease combined with high yield. Recombinant lines in French bean produced from the segregating generation may inherit some degree of resistance if at least one parent is resistant to the anthracnose disease.

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