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Identification of Source of Resistance to Horse Gram Yellow Mosaic Disease (HgYMD) in French Bean (*Phaseolus vulgaris* L.)

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ABSTRACT

Background: Horse gram yellow mosaic disease (HgYMD) is one of the major limiting factor for legumes production in India as it can cause economic loss up to 100%. Till date, horse gram yellow mosaic virus was rare to infect french bean, but now-a-days, it has become the major causal agent for yellow mosaic epidemics in the southern part of India. Few bush type varieties, resistant to yellow mosaic disease are released but not specifically resistant to HgYMD. So, systemic breeding work has been initiated to identify the resistant source for HgYMD in french bean.

Methods: Seventyone genotypes of french bean were screened in a randomized complete block design (RCBD) with three replications under natural epiphytotic as well as artificial condition during Summer, 2018 at ICAR-IIHR, Bengaluru, India.

Result: The genotypes varied for percent disease index (PDI), area under disease progress curve (AUDPC) and the apparent rate of infection per unit time period (r). Two varieties *i.e.*, Jade-5058 and Arka Arjun were showing less PDI of 13.33 and 14.44, least average rate of infection of 0.64 and 0.65 and lowest AUDPC value of 350.54 and 359.16 respectively, compared to other genotypes. Average PDI of Arka Arjun and Jade-5058 were 13.33 and 12.22 under artificial screening, which were much lesser than susceptible check Arka Komal (PDI 92.22) and Arka Anoop (PDI 90.00). Resistant genotypes identified from this experiment can be used for breeding HgYMD resistant french bean lines.

Key words: Disease, French bean, Horse gram yellow mosaic, Resistance, Virus.

INTRODUCTION

French bean (*Phaseolus vulgaris* L., 2n = 2X =22), is one of the most commonly cultivated legume vegetables (also known as snap/garden/string bean), grown for it's tender green pods across different geographical regions of the world. It is rich source of dietary fiber, flavonoid, antioxidants, vitamins, iron, calcium and also acts as laxative, anti-carcinogenic (USDA, 2016). There is good demand and consumer preference of these beans for fresh consumption, cooked or for processing. But, this crop is severely prone to a several viral diseases and among those, horse gram yellow mosaic virus (HgYMV) has become severe in southern India (Muniyappa and Veeresh, 1984) and it causes economic losses up to 100% (Mogali *et al*., 2020). The host range of HgYMV is very rare and specific (Maruthi *et al*., 2006) but Nagaraj (1982) reported that HgYMV can be transmitted to 19 species of leguminosae family and french bean was one of those. Yellow mosaic disease, infecting french bean, produces typical yellow mosaic symptoms (Mishra *et al*., 2020). Although, Aghora *et al*. (2010) reported two french bean lines (IC 525260 and IC 525284) resistant to MYMV, still there is no resistant/tolerant cultivar against HgYMD. Traditionally, the disease management is through vector (white fly) control, but it is not so effective, economical and eco-friendly. So, identification of resistant sources against HgYMD is most suitable disease management strategy which will speed up the resistance breeding and its diversification.

MATERIALS AND METHODS

The experiment was conducted during Feb to May, 2018, when the vector population and natural incidence of HgYMV were high at the experimental plot, ICAR-Indian Institute of Horticultural Research, Hessarghatta, Bengaluru, India.

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Source of virus inoculums and maintenance of virus isolates

Susceptible check cultivars exhibiting typical HgYMV symptoms in the field were collected and planted in earthen pots inside the screen house for maintaining and multiplying disease inoculum. Non-viruliferous whiteflies (*Bemesia tabaci*) were reared and multiplied on eggplant (*Solanum melongena* L.) in screen house. Viruliferous whiteflies were obtained by caging non-viruliferous whiteflies on HgYMV infected plants of susceptible cultivars.

Planting material

Seventy-one genotypes, consisting of germplasms, advanced breeding lines and cultivars from different sources were evaluated for identifying resistance against HgYMD.

The crop was raised according to standard cultivation package and practices. No chemicals have been used to allow disease development to it's full potential. Both natural and artificial screening have been conducted.

Natural screening

All genotypes including susceptible checks (Arka Komal and Arka Anoop) were screened for resistance to LYMD. All bean lines were sown in a randomized complete block design (RCBD) with three replications. To increase the viral inoculum in experimental plot, one row each of HgYMD susceptible checks were planted (Naveen, 2008) at every fifth row in the experimental plot (Infector row technique).

Percent Disease Index

Scoring was done according to Singh *et al*. (2004) (Table 1 and Fig 1).

Per cent disease index has been recorded as,

Per cent disease index =

$$\frac{\text{Sum of numerical value} \times 100}{\text{Number of plants graded} \times \text{Maximum rating}}$$

Apparent rate of infection (r)

Speed, at which an epidemic develops, is called the apparent rate of infection (r). The disease index data was recorded at weekly interval for 5 weeks and used to calculate the apparent rate of disease development using the formula suggested by Van der Plank (1968), where r is the apparent rate of infection in non-logarithmic phase, X_1 and X_2 symbolizes the per cent disease index at time t_1 and subsequent week time t_2.

$$r = 2.3/t_2 - t_1*[\log\{X_2*(1 - X_1)/X_1*(1 - X_2)\}]$$

Area under disease progress curve (AUDPC)

To differentiate between resistant and susceptible genotypes AUDPC is another criterion which calculate the speed of disease progress in plant tissue. Disease index, recorded at seven days interval has been used to calculate the area under disease progress curve (AUDPC) as a measure of quantitative disease resistance. Based on the disease scores, it has been calculated using the following formula (Jeger and Rollinson, 2001),

$$Ak = \sum_{i=1}^{Ni-1} [(y_i + y_{i+1})/2]*(t_{i+1} - t_i)$$

Where

y_i = Proportion of disease on the ith observation.

t_i = Time (days) of observation expressed as days after sowing (DAS).

N = Total number of disease severity readings (PDI) recorded during the experimental period.

Artificial screening

The performance of field evaluated resistant genotypes was confirmed through white fly mediated artificial inoculation of virus. Two highly resistant genotypes from field evaluation and two susceptible checks were evaluated by artificial inoculation in net cage (120 × 120 × 120 cm) covered with nylon mesh. Seeds of each genotypes (8 plants) were sown in peg tray along with susceptible checks. Experiment has been conducted in a Completely Randomised Design with three replications. Mass inoculation (1500-2000 number of whiteflies) has been conducted on ten days old seedlings (two true leaves stage) with a 10-14 hour acquisition access period (AAP) and 24 hour inoculation access period (IAP) at every alternate day till 20 DAS (Raj *et al*. 1989). Nair and Nene (1973) reported that 4-10 whiteflies per test plant can achieve 100% transmission of inoculated plants.

Statistical analysis

All the analysis were performed using SAS (2012) version 9.3 accessed at ICAR-IIHR, Bengaluru.

RESULTS AND DISCUSSION

Screening under natural epiphytotic condition

From the field evaluation, it was observed that PDI values of the 71 genotypes ranged from 13.33 to 75.56 (Table 2). The commercial varieties of french bean Jade-5058 and Arka Arjun were showing resistance with PDI 13.33 and 14.44, respectively. Advance Breeding lines IPS-2, IIHR-(19X34)-9 IPS, IIHR-(19X34)-9 IPS-1, IIHR-(19X34)-9 and variety Slenderette were moderately resistant with PDI of 28.89, 30.00, 27.78, 23.33 and 28.89, respectively. Rest of the genotypes showed susceptible (36 genotypes) and highly susceptible (28 genotypes) reaction to HgYMV. In pole beans, Jyothi (2012) recorded the average yellow mosaic disease incidence of 65.27% in Bengaluru district and the severity of the disease varied from 20.0-88.44%.

Confirmation under artificial conditions

Average PDI of Arka Arjun (13.33) and Jade-5058 (12.22) were much lesser than susceptible check Arka Komal (PDI 92.22) and Arka Anoop (PDI 90.0) (Table 3). Under both

Table 1: Disease scoring scale for Horse Gram Yellow Mosaic Virus in french bean (Singh *et al.*, 2004).

Grade	Description / symptoms	Infected leaf area
1	No visible symptoms on leaves or very small yellow specks with restricted spread.	less than 5% of leaf area
3	Yellow mottling of leaves.	5.1-15 % of leaf area
5	Yellow mottling and discolouration of leaves.	15.1-30% of leaf area
7	Yellowing, pronounced mottling and discolouration as well as foliage reduction	30.1-75 % of leaf area
9	Severe yellowing/entire discolouration of leaves/foliages and pods, stunting of plants, no pod formation or reduction in pod size	above 75 % foliage

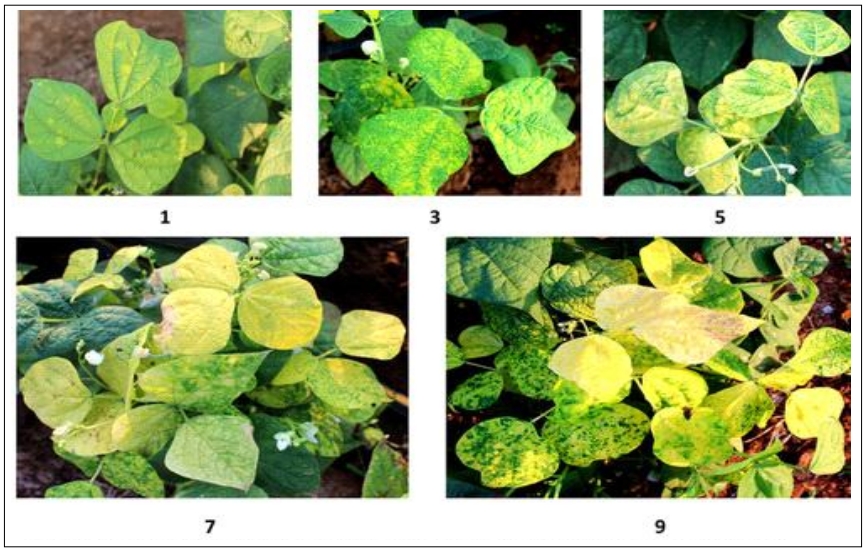

Fig 1: Different grades of disease scoring scale for horse gram yellow mosaic virus in French Bean.

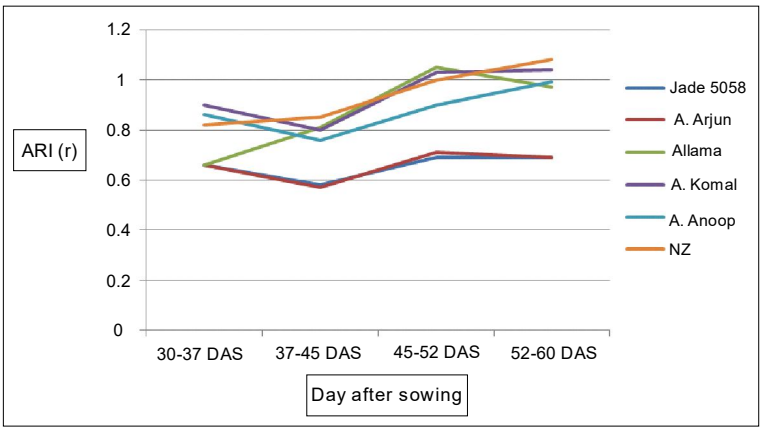

Fig 2: Apparent infection rate of resistant, highly susceptible and susceptible check genotypes.

field and artificial condition, resistant genotypes showed resistance reaction.

Apparent rate of infection (r)

An increase in the infection rate of HgYMV growth was observed in all the genotypes except Arka Arjun and Jade-5058 (Table 4). So that the chances of epidemics is more in almost all the susceptible genotypes of french bean. The maximum average apparent rate of infection value was recorded in the susceptible genotypes Arka Suvidha (1.05), IIHR-(19X34) B (1.03) Arka Bold (1.02) and IIHR-(19X34) 6-1 (1.01). Jade-5058 (0.64) and Arka Arjun (0.65) recorded the minimum 'r' value as compared to susceptible check Arka Komal (0.94) and Arka Anoop (0.88).

The highest rate of disease spread was observed between the fifth and sixth week after sowing of seeds in many genotypes (Fig 2). The apparent rate of infection value varied and during the cropping period they did not remain consistent for aforesaid genotype. The apparent infection rate was highest in most of susceptible genotype but for both resistant genotypes it remained almost constant in between 0.66 to 0.69 through the cropping period. This will help to identify the resistance level with age of plant in all the genotypes.

Disease progression and AUDPC

AUDPC reveals the progress of the pathogen throughout a cropping period (Table 4). Both the resistant genotypes Jade-5058 and Arka Arjun recorded minimum AUDPC value of 350.54 and 359.16 respectively, compared to the other genotypes under screening. The maximum AUDPC value was observed in genotype Arka Suvidha (1363.34) and susceptible check values were 998.89 (Arka Komal) and 776.67 (Arka Anoop).

The difference among resistant, highly susceptible and susceptible check genotypes respect to percent disease index and disease progression was studied in Fig 3. Genotype Arka Arjun and Jade-5058 had lowest disease progression till the end of cropping period (14.44 and 13.33 PDI) compared to two susceptible check Arka Komal and Arka Anoop (66.67 and 54.44 PDI) and highly susceptible genotype NZ and Allamma, which reached to 75.56 and 67.78 PDI, respectively at eighth week. For Allamma,

Table 2: Screening of french bean genotypes for HgYMD resistance.

Genotypes	60 DAS		DR
Jade - 5058	13.33	(21.41)	R
IPS - 2	28.89	(32.50)	MR
Allama	67.78	(55.57)	HS
Arka Sukomol	60.00	(50.77)	HS
US 2	60.00	(50.75)	HS
Arka Arjun	14.44	(22.31)	R
NZ	75.56	(60.37)	HS
Arka Komal	66.67	(54.72)	HS
Arka Anoop	54.44	(47.54)	HS
Arka Sharath	55.56	(48.17)	HS
IIHR- (19X34)-12	50.00	(44.98)	S
IIHR- (19X34) IPS-2	54.44	(47.53)	HS
IIHR- (19X34) 1-4-9	36.67	(37.23)	S
IIHR- (19X34)-1 D.B.	48.89	(44.35)	S
IIHR- (19X34) L.R.	62.22	(52.18)	HS
IIHR- (19X34) IPS 3	42.22	(40.49)	S
IIHR- (19X34) 6-1	55.56	(48.18)	HS
IIHR- (19X34)-1 L.R.	50.00	(44.98)	S
IIHR- (19X34) 1-10 W.S.	51.11	(45.62)	HS
IIHR- (19X34) -1	48.89	(44.34)	S
IIHR- (19X34) 1-5 IPS 3	55.56	(48.18)	HS
IIHR- (19X34) 1-12 IPS 1	41.11	(39.85)	S
IIHR- (19X34) 1-12 L.B.	34.44	(35.90)	S
IIHR- (19X34) W.S.	32.22	(34.55)	S
IIHR- (19X34)-9 IPS	30.00	(33.10)	MR
IIHR- (19X34)-9-IPS 1	27.78	(31.68)	MR
IIHR- (19X34)-9 L.B.	34.44	(35.90)	S
IIHR- (19X34) IPS 1 L.B.	55.56	(48.25)	HS
IIHR- (19X34) 6-1 W	35.56	(36.50)	S
IIHR- (19X34) 1-5 R	35.56	(36.50)	S
IIHR- (19X34) 1-10 W	32.22	(34.55)	S
Markibal 5552	50.00	(44.98)	S
IIHR- (19X34)-12 W	47.78	(43.70)	S
IIHR- (19X34) W	55.56	(48.17)	HS
IIHR- (19X34) B	63.33	(52.73)	HS
IIHR- (19X34)-6 W	64.44	(53.38)	HS
IIHR- (19X34) 9 B	54.44	(47.53)	HS
IIHR- (19X34) 12-1	33.33	(35.15)	S
IIHR- (19X34)-9	23.33	(28.82)	MR
IIHR- (19X34)-12 IPS 4	39.44	(38.89)	S
IIHR- (19X34) 1-5 IPS 4	68.89	(56.13)	HS
IIHR- (19X34)-1 B	46.67	(43.07)	S
IIHR- (19X34) 1-12 IPS 6 L.B.	56.67	(48.82)	HS
Slenderette	28.89	(32.43)	MR
Blue Lake 274	58.89	(50.11)	HS
Crockett	44.44	(41.79)	S
Fantastic Filet	40.00	(39.21)	S
Blue Lake 275	55.56	(48.17)	HS
IIHR-75X55 IPS 1	50.00	(44.98)	S
IIHR- (19X34) 12 L.B.	41.11	(39.80)	S
IIHR- (19X34) -1 D.P.	46.67	(43.07)	S
IIHR- (19X34) 1-12 IPS 6	41.11	(39.83)	S
IIHR- (19X34) D.P.	55.56	(48.17)	HS
IIHR- (19X34) L.B.	47.78	(43.70)	S
IIHR- (19X34) 1-5 IPS 3	41.11	(39.85)	S
IIHR- (19X34) IPS-1	34.44	(35.86)	S
IIHR- (19X34) 6-1 IPS 4	45.56	(42.43)	S
IIHR- (19X34) 1-4 R	46.67	(43.07)	S
IIHR- (19X34) 6-1 IPS 2	41.11	(39.83)	S
IIHR- (19X34) 1-12 W	58.89	(50.11)	HS
IIHR- (19X34)-1 D.P.	57.78	(49.46)	HS
IIHR- (19X34) 1-12-6 W	38.89	(38.55)	S
IIHR- (19X34) 1-5 IPS 1	61.11	(51.44)	HS
IIHR- (19X34)-IPS-1 R	55.56	(48.18)	HS
IIHR- (19X34)-1-5 B	43.33	(41.15)	S
IIHR- (19X34)-1-5 4 IPS	34.44	(35.86)	S
IIHR- (19X34) IPS 2	28.89	(32.35)	S
IIHR- (19X34) IPS 1 W	34.44	(35.90)	S
IIHR- (19X34) 1-5 B	35.56	(36.55)	S
Arka Bold	62.22	(52.46)	HS
Arka Suvidha	64.44	(53.62)	HS
SE m ±		1.750	
C.D.		4.946	

Resistant (R) = 0-15%, Moderately resistant (MR) = 15.1-30%, Susceptible (S) = 30.1-50% and Highly susceptible (HS) = >50%; DAS= Days after sowing; DR = Disease reaction; Data in parenthesis = Angular transformed.

Table 3: Artificial screening of selected genotypes in controlled glass house condition.

Genotypes	PDI %	Transformed values
Arka Arjun	13.33	21.41
Jade-5058	12.22	20.43
Arka Komal	92.22	74.15
Arka Anoop	90.00	71.78
SE m ±		1.76
C.D.		8.20

disease progression started high from fifth week onwards, whereas in Arka Komal and Arka Anoop, it started sixth and seventh week onward. The disease progress curve showed a wide variation between the average disease index for different genotypes evaluated under natural epiphytotic conditions. The variations recorded in the field resistance may be due to the delay in the onset of infection and low speed of disease progression even under strong inoculum pressure (Lebeda, 1999). Disease progress curve reveals the resistance reaction in all the genotypes over the crop growth period (Fig 3). Generally resistant genotypes are having slower disease progression, which usually delays and restricts the pathogen colonization and the expansion of disease symptoms (Mhada *et al.*, 2015).

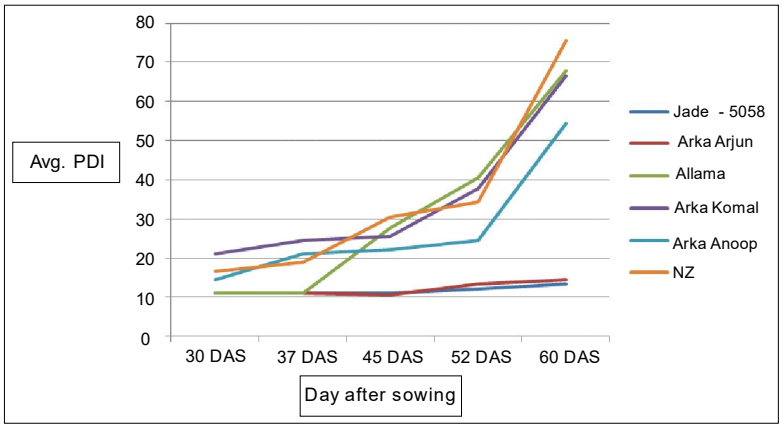

Fig 3: Weekly disease progression of resistant, highly susceptible and susceptible check genotypes.

Table 4: Apparent rate of infection (r) and AUDPC of french bean genotypes for HgYMD.

Genotypes	Average "r"	AUDPC
Jade 5058	0.64	350.54
IPS 2	0.70	421.11
Allama	0.89	905.82
Arka Sukomal	0.78	599.73
US 2	0.73	528.89
Arka Arjun	0.65	359.16
NZ	0.94	989.73
A.Komal	0.94	998.89
A. Anoop	0.88	776.67
A. Sharath	0.86	719.45
(19X34)-12	0.92	913.62
(19X34) IPS-2	0.91	913.61
(19X34) 1-4-9	0.77	535.56
(19X34)-1 D.B.	0.91	892.78
(19X34) L.R.	0.93	948.89
(19X34) IPS 3	0.86	741.11
(19X34) 6-1	1.01	1238.34
(19X34)-1 L.R.	0.90	863.89
(19X34) 1-10 W.S.	0.87	801.67
(19X34) -1	0.91	890.56
(19X34) 1-5 IPS 3	0.91	906.39
(19X34) 1-12 IPS 1	0.87	798.33
(19X34) 1-12 L.B.	0.84	668.34
(19X34) W.S.	0.81	612.22
(19X34)-9 IPS	0.81	621.12
(19X34)-9-IPS 1	0.80	583.33
(19X34)-9 L.B.	0.85	685.00
(19X34) IPS 1 L.B.	0.93	918.33
(19X34) 6-1 W	0.91	856.11
(19X34) 1-5 R	0.85	697.78
(19X34) 1-10 W	0.77	534.45
Markibal 5552	0.88	780.56
(19X34)-12 W	0.95	971.67
(19X34) W	0.97	1072.23
(19X34) B	1.03	1317.23

Table 4: Continue...

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(19X34)-6 W	0.99	1120.83
(19X34) 9 B	0.95	969.17
(19X34) 12-1	0.84	672.23
(19X34)-9	0.76	507.23
(19X34)-12 IPS 4	0.81	617.50
(19X34) 1-5 IPS 4	0.87	822.50
(19X34)-1 B	0.88	795.00
(19X34) 1-12 IPS 6 L.B.	0.86	757.23
Slenderette	0.76	521.11
Blue Lake 274	0.83	682.78
Crockett	0.76	537.22
Fantastic Filet	0.80	582.23
Blue Lake 275	0.78	586.11
75X55 IPS 1	0.83	667.22
(19X34) 12 L.B.	0.87	750.28
(19X34) -1 D.P.	0.86	790.28
(19X34) 1-12 IPS 6	0.86	747.78
(19X34) D.P.	0.99	1167.22
(19X34) L.B.	0.85	746.67
(19X34) 1-5 IPS 3	0.83	671.39
(19X34) IPS-1	0.76	533.06
(19X34) 6-1 IPS 4	0.90	845.00
(19X34) 1-4 R	0.85	696.39
(19X34) 6-1 IPS 2	0.94	943.89
(19X34) 1-12 W	0.95	974.45
(19X34)-1 D.P.	0.99	1136.67
(19X34) 1-12-6 W	0.91	836.11
(19X34) 1-5 IPS 1	0.94	941.67
(19X34)-IPS-1 R	0.95	1018.34
(19X34)-1-5 B	0.80	603.89
(19X34)-1-5 4 IPS	0.78	547.22
(19X34) IPS 2	0.78	550.28
(19X34) IPS 1 W	0.83	647.50
(19X34) 1-5 B	0.82	647.50
Arka Bold	1.02	1296.11
A. Suvidha	1.05	1363.34
SE m ±	0.013	39.74
C.D.	0.036	112.34

In India, there is very negligible research work in french bean against horse gram yellow mosaic virus although it has reports of presence of this virus in french bean long back. Upsurgence of whitefly during last few years further aggravated the YMV situation. The present investigation has implications for breeding YMD resistant french bean lines for the Southern parts of India.

CONCLUSION

Based upon the evaluation under natural and artificial epiphytotic conditions, it can be concluded that the genotypes Arka Arjun and Jade 5058 exhibited resistance with low percent disease indices and progression and can be used for breeding against YMD. High yield, HgYMD resistance and other important agronomic characters can be considered to improve or develop varieties suitable under disease epidemic conditions of Southern India.

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Conflict of interest

All authors declared that there is no conflict of interest.

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