J. Hortl. Sci.
Vol. 16(2) : 301-308, 2021

Original Research Paper

Vegetative vigour, yield and field tolerance to leaf rust in four F_1 hybrids of coffee (*Coffea arabica* L.) in India

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ABSTRACT

Four F_1 hybrids of arabica coffee (*Coffea arabica* L.) developed with the primary objective of pyramiding the S_H3 gene for leaf rust resistance in a commercial variety 'Chandragiri' for achieving the long-lasting resistance to leaf rust, have been evaluated in field. Two hybrids (S.5083 and S.5084) were derived from a donor heterozygous to S_H3, while the other two hybrids (S.5085 and S.5086) were developed from donor homozygous to S_H3. Among the hybrids, S.5086 recorded superior yield performance during individual years with a maximum yield of 1611 kg/ha during 2020-21 and the four year mean yield of 1313 kg/ha. The hybrid exhibited maximum heterosis over mid parent (29.10%) and better parent (17.12%) and S.5086 progeny also manifested high field tolerance to leaf rust pathogen as the entire plant population was free from the disease incidence throughout the study period. The findings of the present study established the efficiency of F_1 breeding strategy with simultaneous pyramiding of rust resistance genes for development of vigorous, high yielding and durable rust resistant F_1 hybrids in arabica. The F_1 hybrid, S.5086 with promising performance in terms of crop yield and high field tolerance to leaf rust has potential implications for commercial exploitation.

Key words: Coffee, F_1 hybrids, durable resistance, pyramiding and S_H3 gene

INTRODUCTION

Coffee, the most popular beverage crop is cultivated in over 50 countries across continents with high export potential. The foreign exchange revenues from coffee exports constitute substantial share in the national economies of the producing countries. The turnover of the world coffee industry is over $ US 40 billion annually and the industry provide direct employment as well as livelihood opportunities to approximately 20 million people in cultivation, on-farm processing, value addition and trade. India is the 7th largest producer of coffee with a cultivated area of 4.59 lakh ha and annual average production of 3.20 lakh MT, which accounts for about 3.5% of the world coffee production and 4.5% share in global exports. Commercial coffee comes from two species viz., *Coffea arabica* L. and *Coffea canephora* Pierre ex A. Froehner that are popularly referred as arabica and robusta coffees, respectively. Arabica coffee types manifest susceptibility to major diseases and pests while robusta is more tolerant to these diseases and pests but the bean and liquor quality of robusta is inferior to arabica. Among the various diseases that affect arabica, coffee leaf rust (CLR) caused by an obligate parasitic fungus, *Hemileia vastatrix* Berk et Br is the most prominent one that cause severe crop losses to an extent of 70% in susceptible cultivars, if proper control measures are not adopted (Anon., 2014). The wet weather with intermittent rainfall & sunshine, relative humidity over 80% and temperatures between 22^0 & 24^0C are the ideal conditions for CLR flare-ups.

Therefore, breeding for rust resistance has been pursued on highest priority in several arabica growing countries including in India, that resulted in development of several coffee varieties manifesting varying levels of resistance to CLR. However, because of the adaptive ability of the *H. vastatrix*, breakdown of resistance under field conditions has been experienced in the commercial varieties due to evolution of new virulent rust races from time to time. At present, as many as 45 different physiological races of rust with ability to infect different coffee genotypes are distributed in various coffee-growing countries (Rodrigues *et al*., 1993 and Prakash *et al*., 2005). The weather conditions that prevail in Indian coffee growing regions are highly favourable for leaf rust pathogen. As a result, high disease build up as well as race mutation leading to the evolution of new virulent races of rust with ability to overcome the resistance in varieties released for cultivation has been a common phenomenon. At present, over 35 different rust races are found distributed in coffee tracts of India. Therefore, development of varieties with long lasting resistance in field is the priority of arabica coffee breeding in India.

The host resistance to coffee leaf rust is reported to be governed by nine resistance genes, designated as S_H1 to S_H9, distributed across the coffee gene pool. Among the commercially cultivated species, four resistance factors (S_H1, S_H2, S_H4, S_H5) were identified in *C. arabica* the only tetraploid species of the genus while four other factors (S_H6, S_H7, S_H8, S_H9), were reported from diploid species, *C. canephora* and S_H3 factor from another diploid species *C. liberica* [Wagner and Bettencourt, 1965 ; Vishveshwara, 1974 ; Bettencourt and Rodrigues, 1988]. It has been well established that the resistance genes identified in *C. arabica*, are less durable under field situations while the genes viz., S_H6, S_H7, S_H8, S_H9 introgressed from the diploid species either from *C. canephora* or from *C. liberica* (S_H3) are found to manifest durable resistance. Hence, the resistance breeding strategies have been mainly focused on pyramiding of resistance genes of diploid origin into selected arabica genotypes by using the natural inter-specific hybrids as donors. In order to expedite the pyramiding of resistance genes into outstanding cultivars by conventional breeding and to reduce the required time frame, F_1 breeding strategy is gaining significance. Further, because of the dominant nature, the resistance genes are expected to express in F_1s resulting in long lasting resistance. With this objective, development of F_1 hybrids has been pursued as a priority of Arabica coffee breeding in India since 2011. The present communication highlights the field performance of four such F_1 hybrids in respect to vegetative vigour, yield and field tolerance to leaf rust.

MATERIALS AND METHODS

The plant material included in the present study comprised of four F_1 progenies, S.5083, S.5084, S.5085 and S.5086 generated from reciprocal crosses between two arabica genotypes, 'Chandragiri' and Sln.10 (Table 1). Two elite plants of Sln.10, characterized as homozygous and heterozygous for S_H3 gene for rust resistance were used in crossing programme, in order to validate the differential response of hybrids, if any. All the four progenies were planted in a compact plot at research farm of Central Coffee Research Institute, Balehonnur, Karnataka, India (13º 22' N, 75º 25' E at an elevation of 2787 MSL), during the year 2012. The weather data pertaining to the study period is furnished in Table 2. In all, 60 plants per progeny were planted in a conventional square design at a spacing of 5'x5' under a two-tier shade canopy, the top cover of evergreen natural forest trees and lower canopy comprising of fast-growing leguminous trees like *Erythrina lithosperma* Miq. (Dadap). The plants were trained on topped single stem system and standard agronomic practices recommended for semi-dwarf arabica genotypes were followed. The progenies have been evaluated for vegetative vigour, field tolerance to leaf rust and crop yield.

Table 1. Details of parents and cross combination of F_1 hybrids

Accession No.	Parents/F_1 cross combination and their response to leaf rust pathogen	
S.3827	Sln.10 (double cross hybrid) {Caturra x Cioccie} x {Caturra x S.795}	Susceptible to 5 races of rust; Selected two plants homozygous and heterozygous to S_H3 gene (donors for S_H3 gene introgressed from a diploid species *C. liberica* and also for good liquor quality attributes)
S.4202	Chandragiri	Resistant Plant selected from the base population that remained tolerant to all prevailing races of rust in India. It's a *C. canephora* introgressed line.
S.5083	Chandragiri	Sln.10 – Heterozygous to S_H3
S.5084	Sln.10 - Heterozygous to S_H3	Chandragiri
S.5085	Chandragiri	Sln.10 – Homozygous to S_H3
S.5086	Sln.10 - Homozygous to S_H3	Chandragiri

Evaluation of agronomic characteristics

For agronomic evaluation in respect of vegetative vigour, data was collected from 20 plants for each progeny (five plants per replication) for three crop seasons till the bush canopy was totally covered. For assessment of vegetative vigour, data on the growth parameters such as stem girth, bush spread, number of primary branches, length of the longest primary, number of nodes per primary, inter nodal length and yield component characters such as number of bearing nodes per primary, number of fruits per node was recorded. The fruit yield per plant and progeny yield was recorded during harvest season i.e., Nov. - Dec. for four crop seasons from 2017 to 2020. Data was also collected from Chandragiri and Sln.10 the parents.

Table 2. Weather data pertaining to the study period (2017-2020)

Months	2017			2018			2019			2020		
	Tem.	Rainfall	RH%	Tem.	Rainfall	RH%	Tem.	Rainfall	RH%	Tem.	Rainfall	RH%
January	22	0 (0)	67	23	0 (0)	81	21	0 (0)	82	23	0(0)	74
February	24	0 (0)	65	24	0 (0)	79	25	0 (0)	62	24	0(0)	61
March	26	13 (0)	65	26	26 (4)	82	25	24 (3)	79	25	23(2)	72
April	27	48 (4)	74	26	108 (16)	78	27	59 (5)	77	26	77(9)	80
May	25	195 (10)	76	25	356 (17)	82	26	39 (6)	87	25	219(14)	84
June	23	404 (23)	85	23	692 (20)	88	25	199 (20)	87	23	270(20)	89
July	22	556 (30)	88	22	1179 (28)	86	24	530 (28)	89	22	440(27)	88
August	23	560 (20)	87	21	1169 (20)	91	23	1204 (31)	86	21	1057(20)	87
September	24	261 (17)	86	23	128 (10)	88	23	538 (21)	88	22	466(17)	88
October	24	109 (6)	87	23	150 (8)	84	24	431(22)	88	22	163(11)	85
November	23	11 (2)	82	23	28 (2)	78	25	48(2)	84	21	44(5)	79
December	22	1 (1)	69	22	25 (1)	78	25	9(1)	85	21	1(1)	85

Tem.-Average temperature, Rainfall - rainy days in parenthesis, RH-Relative Humidity

Data analysis and estimation of heterosis among F_1 hybrids

Analysis of variance was carried out for each character as suggested by Gomez and Gomez (1984). Wherever the treatment differences were found significant, Critical Differences (CD) were worked out at five per cent probability level and values were furnished. The treatment differences that were not significant are indicated as 'NS'.

Relative heterosis and heterobeltiosis manifested by each F_1 hybrid was calculated over mid parent (MPH%) = [(F_1-MP)/MP x 100] and better parent, (BPH%) = [(F_1-BP/BP x 100], respectively.

Evaluation of Coffee Leaf Rust (CLR) incidence:

Observations on leaf rust incidence were recorded during peak periods of disease expression i.e., May-June months during pre-monsoon season and Sept.-Oct. months during post-monsoon season, for four successive years, from 2017 to 2020. Data on CLR incidence was recorded on individual plants and plants with less number of pustules were also treated as susceptible to assess the population susceptible/ resistant in each progeny. The disease build-up on susceptible plants was also scored using the 0-9 scale of Eskes and Toma-Braghini (1981 cf Eskes 1989), where 0 = plants are free from the symptoms; 1= presence of one diseased branch. Likewise, grades were assigned based on progression of disease and 9= maximum disease incidence. Finally, the plants were grouped into four categories viz., 1= tolerant (free from CLR incidence), 2 = moderately tolerant (mild infection without any defoliation); 3= susceptible (medium levels of incidence) and 4= highly susceptible (high disease build up coupled with defoliation).

RESULTS AND DISCUSSION

Agronomic Evaluation – Growth characters

All the four F_1 hybrid genotypes evaluated in the present study exhibited vigorous vegetative growth with compact bush stature which is expected when both the parents are semi-dwarfs. The data in respect of vegetative, yield and yield component characters is presented in Table 3.

Table 3. Character means and analysis of variance for different morphological and yield component characters among different F_1 hybrids

F_1 hybrid/ parental line	Stem girth (cm)	Bush spread (cm)	No. of primary branches	Length of longest primary (cm)	No. of nodes per primary	Internodal length (cm)	No. of bearing nodes/ branch	No. of fruits per node	Avg. fruit yield per plant (kg)
S.5083	38.5	349.5	19.1	98.9	20.0	5.2	9.9	14.7	1.95
S.5084	39.5	366.1	19.1	98.5	20.2	4.9	8.8	14.9	1.78
S.5085	37.0	333.7	19.8	94.3	19.5	4.7	9.1	14.0	1.89
S.5086	36.1	341.2	19.8	94.3	19.2	4.6	8.9	13.7	1.98
Chandragiri (Parent 1)	36.2	322.2	20.0	87.4	19.1	4.9	8.6	12.3	1.69
Sln.10 (Parent 2)	37.3	341.6	21.3	92.3	19.0	5.0	8.7	12.9	1.37
SeM±	1.9	12	0.5	5.3	1.5	0.7	0.6	0.5	0.14
CD @ 5%	NS	NS	NS	NS	NS	NS	NS	NS	NS

From the data it is apparent that the hybrids exhibited uniform vegetative vigour with marginal differences in various characters. Among the F_1 progenies, S.5084 recorded superior growth in terms of stem girth (39.5 cm), number of nodes (20.2), bush spread (366.1); number of fruits per node (14.9). In contrast, S.5086 recorded more compact growth pattern as reflected in most of the growth parameters recorded, compared to other F_1 hybrid progenies (Table 3). Analysis of variance carried out for each character revealed that the character means among different hybrids are significant at $P<0.05$ with their parent populations except number of nodes per primary. These variations in terms of growth characters can be attributed to the growth pattern of the parents and their cross combination used for generation of hybrids. Prakash *et al* (2006) evaluated 17 elite hybrid progenies for growth parameters, field tolerance to rust, clean coffee yield and bean characteristics in a specific agro-climate in India. Analysis of variance of five morphological characters revealed significant differences between the genotypes in respect of four characters (stem girth, bush diameter, length of longest primary and number of primaries per plant) establishing that the genotypes are moderately heterogeneous for plant architecture. Heritability was high (60%) for yield and 64% for field tolerance to leaf rust indicating low genotype x environment interaction for these traits.

Heterosis for crop yield

Data on year wise clean coffee yield among the F_1 progenies and the parents is presented in Table 4. In general, the production trend over the years from 2017-18 to 2020-21 has shown alternate bearing pattern which is common in Arabica coffee. Further, there has been an increase in quantum of yield both in parents and hybrids over the years. All the hybrids recorded higher yields over parents both during the on and off years. Statistical analysis of year wise yields showed significant differences among different genotypes and also the parents (Table 4). During the lean cropping years of 2017-18 and 2019-20, the mean crop yields (clean coffee) among the four hybrid progenies varied from 996 kg/ha (S.5083) to 1102 kg/ha (S.5086) and 1161 kg/ha (S.5085) to 1232 kg/ha (S.5086), respectively. During the high cropping years i.e., 2018-19 and 2020-21, the mean yields ranged from 1233 kg/ha (S.5084) to 1424 kg/ha (S.5083) and 1415 kg/ha (S.5084) to 1611 kg/ha (S.5086). Among the hybrids, S.5086 recorded superior and consistent performance during individual years with a maximum yield of 1611 kg/ha during 2020-21 and the four year mean yield of 1313 kg/ha.

Among the F_1 hybrids, the relative heterosis (MPH%) ranged from 15.83% (S.5084) to 29.10% (S.5086) while heterobeltiosis ranged from 5.08% (S.5084) to 17.12% (S.5086). Among the four F_1 hybrids, S.5086 recorded superior yield performance.

Among the two parental lines, the yield in 'Chandragiri' ranged from 887 kg/ha (2017-18) to 1303kg/ha (2020-21) while in Sln.10, the year wise yields ranged from 741 kg/ha to 1041 kg/ha. Analysis of relative heterosis and heterobeltiosis revealed that all the hybrids exhibited maximum relative heterosis (MPH%) that ranged from 20.7% (S.5084) to 37.5% (S.5086) during the high cropping year, 2020-21 except in S.5085. As regards to heterobeltiosis, all the hybrids except S.5083 recorded high BPH% that ranged from 10.26% (S.5084) to 24.24% (S.5086) during the low cropping year, 2017-18. These differences in relative heterosis and heterobeltiosis, could be attributed to the yield of the parents during the respective years. Among the four F_1 hybrids, S.5086 recorded high BPH% in all the years that ranged from 15.14% (2019-20) to 23.64 (2017-18) except during 2018-19.

Table 4. Year wise clean coffee yields in F_1 hybrids and parental lines

F_1 hybrid progeny/Parents	Clean coffee yield (kg/ha)			
	2017-18	2018-19	2019-20	2020-21
S.5083	996	1424	1179	1557
S.5084	978	1233	1086	1415
S.5085	1069	1325	1161	1457
S.5086	1102	1307	1232	1611
Chandragiri	887	1224	1070	1303
Sln.10	741	987	883	1041
SEm ±	31.17	73.17	55.56	46.70
CD at P = 5%	93.96	220.56	167.48	140.77

Bertrand *et al.* (2005) evaluated the performance of F_1 hybrid plants derived from *C. arabica* for production variables and reported that the F_1 hybrids produced between 22% (trial 1) and 47% (trial 2) more fresh berries than the parental lines in two separate trials. This difference was highly significant for trial

2 (P = 0.00). From the studies on genetic parameters of Timor hybrid derived Arabica genotypes at IAC, Brazil, Mistro *et al*. (2007) reported that the greatest yield gains were achieved when selection was performed based on plot means and years of high yields. It was reported that under normal climate conditions, coffee yields usually increase from the first until the fourth/fifth year. Thereafter, the biennial yield cycles begin, characterized by the alternate high and low yields. From the results of the present study, it is apparent that both the parental lines and the hybrids reflected the alternate bearing behaviour. However, the consistency in production has been recorded in the hybrids in corresponding on and off years of production, alternatively.

Dula (2019) reviewed the heterosis and combining ability studies for yield of *Coffea arabica* varieties in Ethiopia. From the studies conducted by Mesfin and Bayetta (1983), the extent of heterosis for yield was up to 60% over better parent. Out of nine F_1 hybrids, only one hybrid exhibited negative heterosis of -8%. The highest yielding hybrids Melko-CH2 and Ababuna showed 20% and 18% heterosis over the better parent respectively. Given the fact that the genetic distance and combing ability of the parental lines is critical for achieving the maximum extent of heterosis in F_1 hybrids, the heterobeltiosis to the extent of 23.64% over better parent and 37.5% over mid parent for yield in best performing F_1 hybrid (S.5086) in the present study is a significant point to consider for commercial exploitation.

Coffee leaf rust incidence

Data on coffee leaf rust (CLR) incidence among the four F_1 hybrids and parental lines is furnished in Table 5. Among the hybrids, the leaf rust incidence ranged from nil (S.5086) to 51% (S.5083) while in the two parental lines, the CLR incidence ranged from 2.8% to 30% in Chandragiri and 22% to 73% in Sln.10 during different years of study. From the data in respect of individual F_1 hybrids, it is apparent that the two hybrid progenies, S.5083 and S.5084 recorded relatively high susceptibility as 46.1% and 50.9% of population manifested susceptibility during 2019-20, the year that recorded high rust flare up due to favourable weather conditions. The parental lines also recorded maximum susceptibility (30% in Chandragiri and 73% in Sln.10) during the high rust year (2019-20). In contrast, the remaining two F_1 hybrid progenies (S.5085 and S.5086) maintained high levels of field tolerance as the entire population of these two hybrids are free from the rust incidence. The high rust incidence manifested during the 2019-20 could be attributed to the favourable weather conditions i.e., high rain fall coupled with maximum number of rainy days during July and August months as well as ideal temperatures (19^{0}C to 24^{0}C). The quantum of rainfall though higher during July and August 2018, the number of rainy days were low thereby the rust incidence was relatively low compared to 2019. However, inspite of the favourable weather conditions during 2018 and 2019, the disease build up levels were recorded to be very low, in the hybrids S.5083, S.5084 and parent variety 'Chandragiri', the disease build up was characterized by the non-sporulating necrotic spots (group 2), indicating the high levels of tolerance. Further, the high levels of field tolerance to leaf rust in F_1 hybrid progenies (S.5085 and S.5086) could be attributed to the integration of the S_H3 gene in these hybrid populations as the Sln.10 parent used as S_H3 donor is homozygous to S_H3.

In fact, the very objective of the F_1 hybrid breeding programme was to pyramid the maximum number of S_H genes for rust resistance in a proven commercial arabica genotype to ensure long lasting resistance to leaf rust. The resistance in Chandragiri is governed by the S_H genes of tetraploid arabica origin (S_H1,2,4,5) and S_H genes (S_H6, S_H7, S_H8, S_H9) introgressed to *C. arabica* from diploid species, *C. canephora* (robusta coffee). The F_1 hybrids evaluated in the present study were developed with the primary aim of pyramiding of S_H3 gene of *C. liberica* origin in order to improve durability of resistance in Chandragiri. Two plants of Sln10, homozygous and heterozygous to S_H3 gene were consciously selected and used as donor parents (paternal) in crossing programme. Apparently, the two hybrid progenies, S.5083 and S.5084 were derived from the heterozygous donor while the other two hybrid progenies, S.5085 and S.5086 were developed from the homozygous donor plant. Thus, the variability for field tolerance to leaf rust in these four F_1 hybrid progenies, could be attributed to the differences in pyramiding/ integration of S_H3 gene from heterozygous and homozygous donor plants. Analysis of the selected F_1 plants representing the susceptible and resistant plants in different hybrid progenies with SCAR marker linked to S_H3 gene confirmed the presence of the S_H3 gene (un-published data). This inference lend credence from the findings of Shigueoka *et al.* (2014) evaluated nine arabica coffee progenies with an objective to select high-yielding coffee progenies with resistance to coffee leaf rust for the state of Parana in Brazil. It was reported that the plant population of several genotypes derived from 'Sarchimor' and 'Catucai' were susceptible to coffee leaf rust and complete resistance was broken in several coffee plants of 'Catucai' germplasm. However, the interesting observation reported was that the genotype 'F_6 of Catuai x (Catuai x BA-10 coffee)' probably a carrier of S_H3 gene manifested complete resistance in more than 80% plants and inferred that the genotypes were heterozygous to S_H3 gene. Infact, there were several other earlier reports from Brazil that the coffee genotypes carrying S_H3 gene manifested complete resistance to leaf rust (Fazuoli *et al.*, 2005, Pereira *et al.,* 2005. Sera *et al.*, 2007). The findings of the present study also in conformity with that of the earlier reports on high levels of field tolerance manifested by the two F_1 hybrids, S.5086 & S.5085 derived from crosses employing Sln10 plant homozygous to S_H3 gene.

Table 5. Leaf rust incidence (% population) in different F_1 hybrids

Acc. No	2107-18	2018-19	2019-20	2020-21
S.5083	10.3	44.5	50.9	10
S.5084	8.2	12.8	46.1	11
S.5085	0.0	0.0	0.0	5.0
S.5086	0.0	0.0	0.0	0.0
Chandragiri	2.8	20	30	30
Sln.10	22.0	60	73	60

CONCLUSION

The findings of the present study are of high applied value and clearly established the efficiency of F_1 breeding strategy for development of vigorous, high yielding and durable rust resistant F_1 hybrids in arabica. The extent of heterosis in arabica coffee is found to be dependent on the genetic distance and combining ability of the parental genomes. The two F_1 hybrids, S.5086 & S.5085 that recorded promising performance in terms of crop yield coupled with high field tolerance to leaf rust have potential implications for commercial exploitation.

ACKNOWLEDGEMENT

The authors are grateful to the Director of Research, Central Coffee Research Institute, Coffee Board, for the support and encouragement received throughout the research study.

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(Received on 23.05.2021, Revised on 24.07.2021 and Accepted on 27.07.2021)