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Case Study

Breeding for Durable Rust Resistance in Arabica Coffee (*Coffea arabica* L) in India – Implications of Pathogen Diversity

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A B S T R A C T

Coffee is an economically important crop for over 80 countries including India that cultivates on commercial scale and earn substantial foreign exchange through exports. Of the two commercially cultivated coffee types, *Coffea arabica* L. is prone to several diseases and coffee leaf rust known as CLR is the most serious disease-causing enormous crop losses. Though, timely sprays of prophylactic and systemic fungicides effectively control the disease, growing tolerant cultivars has been the most preferred option by the growers. Therefore, breeding for rust resistance has been the main thrust of Arabica coffee breeding across the world more importantly in India as the shade grown cultivation practiced coupled with ideal climatic conditions result in severe devastation. The article highlights the successes and disappointments of the systematic breeding programmes undertaken at Central Coffee Research Institute, especially the introgressive breeding. The implications of the vast diversity of the CLR pathogen, *Hemiliea vastatrix*, an obligate parasite on the longevity of rust resistance in the CLR resistant varieties released for cultivation. The gene pyramiding strategy for achieving durable rust resistance and integration of marker assisted selection for improving the selection efficiency are detailed and discussed.

Key words: Coffee leaf rust, Arabica coffee, Resistance breeding, Pathogen diversity, Durable resistance

Coffee is one of the commercially important crops in tropics and sub-tropics and popularly referred as 'Brown Gold' because the economy of several producer countries depends on exports earnings of this commodity. Coffee attracts global attention as it is largely cultivated in developing countries but traded to developed countries, the major consumers of this beverage. The entire coffee production of the world comes from only two species, *C. arabica*, called as Arabica coffee and *C. canephora* Pierre ex Froehner commonly known as Robusta coffee. India grows both these commercial coffee types depending on their adoptability to high and low altitudes, respectively covering an area of 4.59 lakh ha and produces around 3.19MT annually with 4.5% share in global production [1]. Coffee cultivation in India is unique under two-tier shade canopy while in all other major coffee producing countries, coffee is grown in open conditions without any shade.

Arabica coffee is susceptible for many diseases of which, coffee leaf rust (CLR) caused by a fungal pathogen *Hemileia vastatrix* is the most serious one causing severe crop losses in almost all Arabica coffee growing countries. In India, CLR is a great threat to arabica cultivation because the climatic factors are favourable for high disease build up, leading to crop losses up to 70% in susceptible cultivars, if proper control measures are not adopted [2]. Coffee leaf rust (CLR) is one of the classical plant diseases that was though considered to have originated on wild coffee types in Ethiopia [3] the first published report on cultivated coffee was from Sri Lanka in 1869 [4]. The disease gradually spread to other Asian countries like India and Indonesia soon but to majority of the coffee growing countries by late 19th century. The most prevalent management practices include the prophylactic sprays of Bordeaux mixture or specific systemic fungicides. Nevertheless, growing disease tolerant varieties is the best option preferred by the coffee farmers. Therefore, breeding for rust resistance has been the important objective of the Arabica coffee improvement across the Arabica growing countries in the world, including India. The breeding programmes undertaken in different countries were successful in development of several resistant varieties for commercial cultivation. But, quite often, there were several disappointments reported across the countries because of the breakdown of resistance in the commercial varieties which was primarily due to evolution of new virulent races. Therefore, breeding for durable rust resistance is the current priority that is being pursued by integration of conventional breeding methods with advanced strategies. Therefore, the present paper highlights a comprehensive picture on leaf rust resistance breeding in India and recent initiatives towards achieving durability of resistance in improved coffee varieties released for commercial cultivation in the country.

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Commercial coffee types, breeding behavior and resistance spectra

Though the genus *Coffea* represent over 100 species, the commercial coffee production relies on two species, *Coffea arabica* L and *Coffea canephora* Pierre ex Froehner. *C. arabica*, popularly known as arabica coffee is an allo-tetraploid and self-compatible species. On the other hand, all other species including *C. canephora*, which is referred as robusta coffee are diploids, self-incompatible and strictly cross pollinated. Arabica is susceptible to major diseases and pests like leaf rust (*Hemileia vastatrix* Berk & Br) (Fig 1), Coffee berry disease (*Colletotrichum kahawae* Waller et Bridge), stem borer (*Xylotrechus quadripes* Chevrolat) and nematodes (*Meloidogyne* sp. and *Pratylenchus* sp.) while Robusta is more tolerant to these diseases and pests. As regards to beverage quality, Arabica produces superior quality coffee compared to Robusta. Resistance to coffee leaf rust in various coffee species is reported to be determined by at least nine resistance genes, S_H1 to S_H9, either singly or in combination. The corresponding virulence factors in the pathogen are designated as v1 to v9. Of these resistance genes, S_H1, S_H2, S_H4 and S_H5 were identified in the tetraploid species *C. arabica* while S_H6, S_H7, S_H8 and S_H9, were reported from the diploid species *C. canephora*. The S_H3 gene was introgressed from another diploid species *C. liberica* [5-7].

Fig 1 Coffee leaf rust disease on Arabica coffee

Pathogen diversity and behaviour of resistance genes in field situations

Like any other rust fungi, coffee leaf rust fungus does possess the ability to exist in different physiological forms popularly called as races. The pioneering work on existence of physiological races of coffee leaf rust pathogen, carried out in India by W.W. Mayne, during 1930s is a classical work on CLR. This pioneering work resulted in identification of four races of the rust fungus, viz. race I & race II [8] race III [9] and race IV [10]. At present, 45 physiological races of rust with virulence to infect various coffee genotypes have been characterized from samples collected across the coffee-growing countries [11-12]. Out of these characterized race types, 37 races are found distributed in India [13]. Because of such wide rust race diversity, rust resistance breeding programmes resulted in successes as well as disappointments due to appearance of new virulent Races of rust fungus from time to time [14]. Performance of the varieties possessing different resistance genes in field conditions established that the resistance genes identified in *C. arabica* (S_H1, S_H2, S_H4, S_H5) present either singly or in combination, did not provide long lasting resistance to most of the races of rust fungus. In contrast, the genes introgressed from the diploid species such as the S_H3 gene from *C. liberica* and also the genes from *C. canephora* (S_H6 to S_H9) are found to provide durable protection to arabica, as compared to the genes identified in tetraploid *C. arabica* itself [15].

Breeding for rust resistance in India – An over view

The very objective of initiating organized research on coffee in India during 1925 was breeding rust resistant varieties and to find solutions for rust problem that started devastating arabica coffee during late 1870s. Thus, systematic breeding programmes undertaken over the years resulted in development of 13 varieties of arabica that manifest varying levels of resistance under field conditions. In early phase of coffee breeding carried out during the period from 1925 to 1940s, a natural inter-specific hybrid (*C. arabica* and *C. liberica*) derivative, S.26 was used as donor for rust resistance and two arabica varieties viz. S.288 and S.795 were developed. The S.795 became very popular with yield potential of 1500 kg/ha and wider adaptability [16]. Resistance to leaf rust in these two varieties (S.288 and S.795) was governed by S_H3 resistance factor for coffee rust, introgressed from *Coffea liberica,* through the spontaneous hybrid S.26. This introgressed gene (S_H3) is mainly responsible for imparting rust resistance in S.795 for several years in field till the origin of more virulent races.

In the second phase of breeding during the period from 1950s to 1980s, the spontaneous hybrids of *C. arabica* and *C. canephora* such as 'Devamachy' identified in India and Hibrido de Timor spotted in Timor island of Indonesia that was introduced to India from Coffee Rusts Research Centre (CIFC), Portugal which is popularly known as (HDT – CIFC 1343) were exploited in breeding as resistance donors. Thus, in this second phase of breeding, the *C. canephora* (Robusta) introgressed lines were used and four varieties viz., Sln.5A, Sln.5B, Sln.8 and Sln.9 were developed. All these varieties have been recommended for commercial cultivation depending on their location specific suitability. These tetraplod inter-specific hybrid derivatives manifest high field tolerance to rust with a production potential of 1200 to 1600 kg/ha depending on the cultivation practices and growing conditions. The success in introgressive breeding formed the basis for developing another variety, Sln.6 by crossing *C. arabica* and *C. canephora* followed by recurrent back crossing to arabica parent. This variety is known to possess arabica phenotype with resistance and crop bearing characters of robusta. The bushes of Sln.6 are tall, vigorous with profuse branching pattern and yield potential of 1200 – 1500 kg/ha. The Sln.6 variety is suitable for cultivation in transitional altitudes (900 m to 1000 m) and 80% of plant population manifests high field tolerance to rust.

In subsequent phase of breeding during the period from 1980s to 2000s, the rust resistance breeding was pursued on exploitation of dwarf mutants viz., San Ramon, Caturra, Villasarchi with the main objective of developing semi-dwarf varieties with compact bush stature suitable for high density planting. As these dwarf mutants were identified in *C. arabica* populations, though possess good production potential, manifest high susceptibility to leaf rust. Therefore, few HDT plants (spontaneous Robusta x Arabica) collected by CIFC, Portugal in Timor island of Indonesia (HDT CIFC 832/1, 832/2 and 1343) were used as resistance donors four dwarf/semi-dwarf varieties viz., San Ramon hybrids, Sln.10, Cauvery/Catimor and Chandragiri/Sarchimor were developed for commercial cultivation. The San Ramon hybrids were derived from multiple crosses between `San Ramon`, a dwarf mutant and other tall arabicas like S.795, Agaro and Hibrido de Timor (HDT). The San Ramon hybrids (Sln.7) segregate for phenotype (dwarf, intermediate and talls) and dwarf phenotypes with yield potential of over 1500kg/ha are suitable for high density planting (4ft x 4ft, 5ft x 4ft.). The Sln.7.3 is a late ripener, drought tolerant and suitable for marginal areas. The Cauvery/Catimor was derived from cross between Caturra a high yielding dwarf mutant with HDT (CIFC-832/1) variety that was given for commercial cultivation in India during 1980s. The variety became popular because of its high yield potential of 1500-2000 kg/ha and resistance to leaf rust in initial years. However, within few years, the variety manifested susceptibility to new virulent races of rust, indicating the less durable nature of resistance donor, HDT, CIFC- 832/1. Rust disease management became critical for realizing economic yields and hence cultivation of this variety is now limited to high elevations. Later, `Chandragiri` derived from cross between Villasarchi x HDT (CIFC-832/2) popularly known as Sarchimor was released for commercial cultivation during 2007-08. This variety with a yield potential of 1500 to 1800 kg/ha coupled with high field tolerance to leaf rust has become popular because of consistent performance with respect to crop yield, field tolerance to leaf rust and value addition in terms of bold beans. Thus, the successes achieved in rust resistance breeding in India helped to a great extent in managing the coffee leaf rust in field conditions. Growing the leaf rust tolerant cultivars integrated with prophylactic sprays of Bordeaux mixture has been successfully adopted by the coffee growers.

Breeding for rust resistance and origin of rust races in Indian context

In general, the origin and distribution of rust races depends on the resistance factors present in corresponding coffee genotypes distributed in field. This was apparent from characterization of rust samples received at CIFC from different coffee growing countries and also periodical surveys carried out in countries like India, Ethiopia, and Brazil [17-18]. In India, the origin and distribution of rust races has been in concurrence with the resistance factors of the coffee genotypes under cultivation.

In the long pursuit of rust resistance breeding in India, all the nine resistance factors i.e., S_H1 to S_H9 identified so far, have been exploited in evolving rust resistant varieties for commercial cultivation. Initially, resistance factors of arabica (S_H1, S_H2, S_H4 and S_H5) and S_H3 from *C. liberica* were exploited. This has resulted in origin of rust races with corresponding virulence genes I (v2,5), II (v5), VIII (v2,3,5), XXIII (v1,2,4,5) and XXIV (v2,4,5). From the initial experience, it was found that integration of resistance genes of arabica origin provided short-lived respite and only S_H3 a gene from a diploid species, *C. liberica* provided long lasting resistance. This paved the way for experimenting with the other genes of resistance found in other diploid species, especially *C. canephora*. In order to exploit the rust resistance genes, present in *C. canephora*, inter-specific hybrids between *C. canephora* and *C. arabica* were developed and Sln.6 was evolved. Further, several other varieties like Sln.5A, Sln.5B, Sln.8, Sln.9 were also developed using the spontaneous hybrids of Robusta x Arabica like Hibrido de Timor (HDT) and Devamachy. Periodic sampling and characterization of rust spores on some of these genotypes established that new virulent races, XXIV (v2, 4,5,) and XXV (v2,5,6) were found occurring on Robusta x Arabica hybrids and XXV on HDT descendants [19]. Thus, origin of a new rust race XXV in India consequent to the release of Robusta x Arabica hybrids reflects the adaptive ability of the rust pathogen. Subsequently, semi-dwarf varieties like, Cauvery/Catimor and Sln.10 were developed by crossing the dwarf mutants with Hibrido de Timor (HDT) as resistance donor parent. Within six to seven years after the release of Cauvery/Catimor variety for commercial cultivation, four new rust races with new virulent gene combinations (v2,5,6,7,9; v2,4,5,8,?; v2,5,6,7,9,?; v2,4,5,6,7,8,9) were characterized at CIFC [20], from the rust samples collected on Cauvery/Catimor genotypes. Thus, the appearance of new virulent rust races with new gene combinations has always been in correspondence with the resistance factors (S_H1 to S_H9) incorporated into new coffee varieties released for commercial cultivation, leading to breakdown of resistance in newly evolved materials.

Implications of resistance breeding on host – pathogen interactions

From the observations on interactions between the development of the new varieties with improved rust resistance and evolution of new virulent rust races, it could be inferred that cultivation of diverse rust resistant varieties in field exerted selection pressure on rust pathogen. As a consequence, the pathogen is forced to mutate by acquiring new virulence genes with ability to breakdown the resistance of the improved/resistant varieties. This phenomenon is more frequent in India because of the ideal weather conditions prevailing in Indian coffee tracts that facilitate quick mutations in rust pathogen. This inference lends credence from the fact that out of 45 different physiologic races of coffee leaf rust isolated/characterized so far across different coffee growing countries, ~ 37 races are known to be distributed in India [21]. The new virulent races during the process of evolution acquired the adaptive ability to overcome the resistance of the improved varieties in favorable conditions. Nevertheless, close monitoring the distribution of virulent races in field situations revealed that, though new virulent rust races with eight virulence genes have been identified, the distribution of rust races is in accordance with the spread/distribution of host plant material and also at the cost of fitness penalty.

Durability of resistance as influenced by pathogen diversity

From the Indian experience, it has been well established that though remarkable success has been achieved in breeding rust resistant/tolerant varieties for commercial cultivation, durability of resistance/tolerance in these varieties has been influenced by the concurrent increase in pathogen diversity. In the back drop of increased pathogen diversity various donors of resistance such as HDT lines have been continuously monitored in Indian conditions. Interestingly, the HDT CIFC 832/1 is found less durable compared to HDT - CIFC 832/2 and HDT - CIFC 1343 which provided important insights for selection of appropriate strategies for breeding for long lasting resistance/tolerance to leaf rust pathogen in spite of its vast diversity.

Current focus of breeding in India to achieve durable rust resistance

It is an established fact that even though the adaptive capacity of the *H. vastatrix* with ability to overcome host resistances has been gradually increased the resistance genes introgressed from the diploid species such as the S_H3 gene from *C. liberica* and S_H genes from *C. canephora* (S_H6 to S_H9) could sustain pathogen virulence with complex interactions.

Thus, these genes of diploid origin could be able to provide long - lasting protection in certain genotypes of arabica under field conditions, as compared to the genes identified in tetraploid *C. arabica*. However, considering the adaptive ability of the pathogen, breeding for rust resistance is a continuous process. Therefore, some of the promising approaches to achieve durable rust resistance include pyramiding of resistance genes of diploid species in a selected arabica genotype or integrated use of these genes in composite varieties. Nevertheless, combined introgression of various genes of resistance into outstanding cultivars by conventional breeding is a challenging task in a reasonable time-frame. In addition, characterization of the resistance spectra of individual genotypes by following a series of inoculations on selected rust differentials/indicator plants is a very laborious and time taking process. In this context, development of molecular markers linked to S_H genes and integration of marker assisted selection/tracking of resistance genes would not only expedite the breeding but also increase the efficiency of selection.

Accordingly, the current focus of arabica coffee breeding in India is on development and evaluation of reciprocal hybrids of *C. liberica* introgressed lines and robusta introgressed lines, derived from HDT 1343 and HDT 832/2. This gene pyramiding approach is expected to integrate all the rust resistance genes of diploid species in tetraploid arabica background that ensure durable and long-lasting resistance to leaf rust. In this direction, several hybrid progenies have been developed between Catimor genotypes and liberica introgressed lines. Field evaluation of these hybrids resulted in selection of two promising progenies (S.4814 and S.4817) that recorded superior performance for yield, field tolerance to rust, bean and liquor quality traits [22]. Furthermore, four progenies (S.5083 to S.5086) have been developed from the crosses between Sarchimor genotypes and *C. liberica* introgressed lines and being evaluated at Central Coffee Research Institute (Fig 2). While all these genotypes are semi-dwarf phenotypes, two more hybrid progenies of tall phenotype (S.5091 & S.5092) were also established from crosses involving tall phenotypes S.2790 and S.2724. The breeding protocol has been hastened by integration of marker assisted selection with SCAR markers linked to S_H3 gene for tracking S_H3 gene in parental lines as well as hybrid populations. From preliminary evaluation, the field performance of these genotypes is found promising both in respect of all agronomical traits and field tolerance to leaf rust, which provide a way forward towards breeding for durable rust resistance in arabica coffee.

Fig 2 Leaf rust tolerant Arabica genotype – S.5085

CONCLUSIONS

Resistance management is one of the most critical criteria in devising pragmatic strategies for management of plant diseases and more particularly the classical diseases like coffee leaf rust caused by obligate parasitic fungal pathogens. For success in rust resistance breeding, understanding the resistance sources available in coffee gene pool, their resistance spectra, pathogen diversity and behaviour of resistance genes in field situations are particularly important. In this context, the article covering an overview of rust resistance breeding in India clearly highlighted the host-pathogen interactions, origin of rust races in Indian context and durability of resistance as influenced by pathogen diversity. In the backdrop of these developments and by considering the complexity of coffee leaf rust pathogen, the current focus of breeding in India on pyramiding of resistance genes looks promising towards achieving durable rust resistance in commercial coffee varieties.

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