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Status and strategies in breeding for rust resistance in wheat

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ABSTRACT

Wheat along with rice and maize is fulfilling half of the calories demands of the world. Global Wheat production has increased tremendously since green revolution in 1960's and helped in minimizing hunger and malnutrition. Developing countries, which consume 60% of the global wheat production, have shown a higher yield increase than the developed countries in the past [1]. It was driven by the hunger prevalence in these countries and was attributable to the introduction of high yielding and rusted resistant semi dwarf varieties developed under the collaborative efforts of International and National research systems during the last 50 years. Whereas, climate change and the emergence of new pests and diseases are threatening the food sustainability. The evolution of new races of disease pathogens like stem rust (Ug 99) is of serious concern. In order to feed the ever increasing population we have to increase wheat production at the rate 1.6% which can be achieved by developing high yielding varieties having a good tolerance level for biotic and abiotic stresses.

Keywords: Leaf Rust; Strip Rust; Stem Rust; Resistance; Wheat

1. INTRODUCTION

Among the most important diseases in wheat that significantly reduce wheat production are those caused by the rusts (leaf, stem, and stripe). The rusts of wheat are among the most important plant widespread pathogens that can be found in most areas of the world where wheat is grown. Wheat leaf rust is caused by *Puccinia triticina* Eriks, wheat stem rust by *Puccinia graminis* f. sp. *tritici*, and wheat stripe rust by *Puccinia striiformis* f. sp. *tritici*. Leaf rust occurs more regularly and in more world-wide regions than stem rust or stripe rust of wheat. Yield losses in wheat from *P. triticina* infections are usually the result of decreased number of kernels per head and lower kernel weight and it may reach 40% in susceptible cultivars [2]. Yield losses caused by the stem rust pathogens in the mid of the 20th century reached 20% - 30% in Eastern and Central Europe and many other countries including Australia, China and India [3]. The yield losses caused by the most virulent stem rust race Ug99 emerged first in Uganda and after that in Kenya, Ethiopia, Yemen, in the Middle East and South Asia and losses were estimated to be approximately USD $3 billions (http://www.seedquest.com).

The disease-causing wheat rust fungi are spread in the form of clonally produced dikaryotic urediniospores, which can be dispersed by wind for thousands of kilometers from initial infection sites across different continents and oceans. Epidemics of wheat rusts can occur on a continental scale due to the widespread dispersal of urediniospores [4]. Wheat rust fungi are highly specific obligate parasites. Their avirulent genes interact with resistance genes in wheat in a gene-for-gene manner. Rust populations can be characterized by distribution of races and the frequencies of virulence against specific rust resistance genes on a defined set of wheat differential hosts. The avirulence genes that are present reflect only a small proportion of the total genetic variation found in rust populations, but this variation is subject to intense selection by the resistance genes in commonly grown wheat cultivars. Selectively neutral markers such as isozymes or more recently developed molecular markers, such as random amplified polymorphic DNA (RAPD), simple sequence repeat (SSR) and amplified fragment length polymorphism (AFLP) can also be used to characterize and compare rust populations. As the wheat rust fungi are spread easily within and between continents, it is essential to document the genetic changes in rust populations over large geographic areas in order to facilitate the development of rational strategies or durable resistance.

2. BREEDING FOR RUST RESISTANCE

The semi dwarf and dwarf varieties developed at CIMMYT, Mexico in the early days of green revolution (Penjamo 62, Pitic 62, Lerma Rojo 64, Sanora 64 and Siete Cerros etc.) had been responsible for yield breakthrough in India, Pakistan, Turkey, Afghanistan and many other parts of the world. The life time of most of these Mexican varieties was short as appearance of new stem rust race has terminated their useful life time however, there were some exceptions also. The variety Lerma Rojo 64 had life time of eleven year, while others like Yaqui 50, Champingo 52 and Champingo 53 retained their resistance until they were displaced from commercial cultivation by new high yielding varieties [5]. The long life of these varieties is attributable to their genetic background. They had combination of Hope and Thacher type and Kenya type resistance. During the period 1965-1985, the CMMYT breeding program has incorporated diversity of genes. Most of the material distributed during this period contains *Sr*2 and two to four additional genes for stem rust resistant. These additional genes include *Sr*5, *Sr*6, *Sr*7a, *Sr*7b, *Sr*8a, *Sr*9b, *Sr*9d, *Sr*9e, *Sr*9g, *Sr*10, *Sr*11, *Sr*12, *Sr*17, *Sr*24, *Sr*26, *Sr*30, *Sr*31, *Sr*36 [6]. The parallel strategy was also adopted by many national programs.

The importance of *Lr*13 gene for leaf rust (*Puccinia triticina*) resistance was recognized in the early 1970's when it was transferred along with other genes in to many wheat varieties. Some varieties containing *Lr*13 in combination with other genes developed in Mexico, India and Pakistan are given in **Table 1**. The gene, *Lr*13 itself does not provide desired level of resistance but when present in combination with other genes it provides a degree of resistance of high probability of being durable. The mode of action of *Lr*13 complex in CIMMYT program is non specific type of resistance. Its presence in combination with *Lr*34 in some members of Bluebird series gave them long life. Another example of this combination is a Pakistani variety *Lyalpur* 73 which although replaced in the farmers field by the introduction of new high yielding varieties but even after 36 years of release, it still have very good resistance for leaf rust (20 M) in screening nurseries. The varieties like *Genero*81 and *Torim* 73 which remained resistance to leaf rust in Mexico for long time also have *Lr*34 gene in combination with other genes.

The adult plant resistance to leaf rust of the Brazilian wheat cultivar "Frontana" was first described as due to the gene *Lr*13 (effective in the adult plant stage) and one or two modifiers [7].

Subsequent studies by [8] indicated the presence of *Lr*34 and *LrT*3 in "Frontana". Reference [9] showed that

Table 1. Leaf rust resistance genes in old wheat varieties.

S No	Varieties	Year	Country/Region	Genes
1	Lerma Rojo	1964	Mexico	*Lr*13, *Lr*17
2	Champingo 53	1953	Mexico	*Lr*34
3	Penjamo 62	1962	Mexico	*Lr*14*a*, *Lr*34
4	Pitic 62	1962	Mexico	*Lr*14*a*
5	Sonora 64	1964	Mexico	*Lr*1
6	Mexipak 65, Kalyansona	1965	India, Pakistan	*Lr*14*a*
7	Sonalika, Bluesilver	1967, 1971	India, Pakistan	*Lr*13, *Lr*14*a*
8	Lyalpur 73	1973	Pakistan	*Lr*1, *Lr*13, *Lr*34
9	Bluebird, Yecora 70	1970,s	Mexico, Pakistan	*Lr*1, *Lr*13, *Lr*34
10	Ciano 79	1979	Mexico	*Lr*16
11	Arz	1973	Lebnon	*Lr*17
12	Pavon F 76, Dollarbird	1983, 1987	Mexico, Australia	*Lr*1, *Lr*10, *Lr*13, *Lr*46+
13	Parula	1981	CIMMYT	*Lr*34 & *Lr*46+
14	Punjab 81	1981	Pakistan	*Lr*10, *Lr*13, *Lr*34
15	Era	1970	North America	*Lr*10, *Lr*13, *Lr*34

*Lr*13 could confer low seedling reaction at elevated temperature. The gene *Lr*13 appears to be common in cultivars from Australia (Hawthorn 1984); India [10]; and Brazil, Argentina and Unites States [4]; and in cultivars derived from CIMMYT germplasm [11]. Gene *Lr*13 has been shown to interact with other genes such as *Lr*16 and *Lr*34 [12]. Durable resistance to leaf rust in various cultivars has been thought to be due to the interaction of *Lr*13 with other *Lr* genes [4]. Reference [13] investigated the inheritance of adult plant resistance in "Frontana" and 3 globally leaf rust resistant CIMMYT spring bread wheat varieties, the genetic test for the presence of *Lr*34, the postulation of the other known *Lr* genes, and the role of *Lr*13 and other known *Lr* genes in conferring adult plant resistance. They concluded that resistance was independent of major genes.

The material developed during mid 1960's had acquired resistance for yellow rust from Andean region varieties which possessed high level of resistance. The Anza was derived from cross LR/N10B//3*ANE and released in North Africa, Sudan, South Africa and New Zealand. It was regarded as durable resistant for yellow rust by reference [14] and may have derived durable resistance from Anderson [15]. Durable resistance of Anza is widely deployed in spring wheat and in some winter wheat varieties. This durable resistance was attributed to the presence of gene Yr18 by reference [16]. The gene, Yr7 is also present in a range of spring wheat and winter wheat varieties and it is frequently associated with Sr9g. It is reported in number of varieties such as Barani 83, PBW12, WL2265, Seri 82 (*Yr*2, *Yr*7, *Yr*9), Pavon76 (*Yr*6, *Yr*7, *Yr*29), Pak.81 (*Yr*7, *Yr*9) [17]. Veery and Pavon containing *Yr*7 have been released in 31 and 16 countries, repectively with different names which show the wide use of *Yr*7 gene.

3. 1B-1R TRANSLOCATION

Reference [18] produced several lines having a translocation between a segment of hairy neck chromosome of rye 5R and different wheat chromosome segments. Due to genetic relationship between rye chromosome 5R and wheat chromosome of homeology group 1, the designation 1R was proposed for this chromosome [19]. The rye chromosome 1R was reported to be containing powdery mildew and stripe rust resistance genes in its short arm. It was found that these genes were linked with stem rust and leaf rust resistant genes and cultivars Salzmunder, Baertweizen and Weique have identical genes [20]. A sister line of these "Neuzutch" was used for breeding in Soviet Union and gave rise to Russian cultivars Kavkaz, Aurora, Besostaya 2, Skorospelka and many others. Neuzutch possesses a complete 1R chromosome, whereas Kavkaz and Aroura have an interchange chromosome having 1B segment and a rye chromosome 1R segment. Kavkaz was introduced in to CIMMYT germplasm where a high yielding spring wheat cultivar "Veery" was released. This segment was also transferred to several Europian cultivars. These cultivars were found possessing resistance to wheat streak mosaic virus and its vector wheat curl mites. There was good compensation of Rye chromosome 1R for the elimination of wheat chromosome 1B. The 1B.1R translocation appears to be more stable and superior in agronomic properties. It was easy for the breeders to work with this translocation as there was no cytological problem associated with it [21]. Therefore, this translocation became widespread in wheat cultivars released in China and USA, India, Pakistan and several other countries during the mid-1980s and later. The Veery derivatives due to their superior agronomic feature and disease resistance were widely cultivated in different parts of the world. This germplasm showed significant grain yield advantage and wide adaptation with superior disease resistance attributes due to the presence of the 1B.1R translocation. The higher yielding ability of 1B-1R germplasm was attributed to post anthesis stress tolerance of this material resulting in higher Kernal weight [22]. The frequency of 1B.1R translocation went up to approximately 70% at one stage in CIMMYT's spring wheat germplasm but has declined to about 30% in more recent advanced lines [23]. This translocation, derived from imperial Rye carries genes *Sr*31, *Lr*26, *Yr*9 and *Pm*8 [24], when used initially it provided resistance to stem rust, leaf rust and yellow rust but with the development of new virulent races, these genes are in effective now [23]. Despite, its successful use it was not widely deployed in Australia due to sticky dough, poor mixing characteristics resulting poor bread making qualities [24].

4. EMERGENCE OF NEW RUST RACES

The wide spread global popularity of the germplasm with 1B-1R translocation created monoculture situation. This lead to the evolution of some new devastating rust races results a serious threat to global wheat production. A race of *P. striiformis*, *Yr*9 was 1st observed in East Africa in 1986 and subsequently migrated to North Africa and South Asia. Once it appeared in Yemen in 1991 it took just four years to reach wheat fields of south Asia [25]. On its way it caused major yield losses in Egypt, Syria, Turkey, Iran, Iraq, Afghanistan and Pakistan exceeding USD 1 billion. Similarly, *Yr*27 emergence and its movement following the same pathway posed major threat to wheat production in India and Pakistan, where mega cultivars PBW343 and Inqilab 91 were having *Yr*27 gene based resistance. In 2005, the wheat crop in Nothern Pakistan was severely hit by this race of yellow rust where most of the area was under Inqilab-91. Stem rust resistance in wheat cultivars with Sr31 remained effective for more than 30 years. In 1990's, most of the wheat varieties were having 1B-1R translocation which created a monoculture situation in Africa, Asia and other parts of the world. Isolates of *Puccinia graminis tritici* (*pgt*), which were virulent on *Sr*31 were collected for the 1st time in Uganda during 1999 and then spread throughout East Africa [26]. It subsequently spread to Kenya and Ethiopia in 2005 [27]. The race, named as TTKS (Ug99), is virulent on majority of mega wheat varieties and can cause 100% yield losses whereas, up to 80% yield losses have been reported in Kenya. A new variant of this stem rust race has been found in Kenya since 2006, which is virulent on Sr24 [28,29]. Now a day fungicides are being used to control stem rust in Kenya [30]. It ultimately jumped the red sea and its presence has been reported in Yemen since 2006 and was also found in Sudan in the same year. In March 2007, isolates of *pgt* were collected from different locations in Iran and the collections from Borujerd and Hamadan were identified as TTKSK [31]. The race identified, produced high IT's of 3 to 4 on wheat genotypes carrying 1BL-1Rs translocation (Falat and PBW343). Subsequently, FAO announced its existence in Iran and alarmed a threat for, the breadbasket zone of the world, South Asia and other neighboring regions. A new race of stem rust virulent on *Sr*25 gene, have been detected in India [32]. This isolate collected from Karnataka, has shown IT's 3+ to 4 on primary leaves of differential types with *Sr*25 gene. This race is named as PKTSC according to North American system. The detection of *Sr*25 virulent race alarmed the breeders that they should breed for adult plant resistance or pyramid 2 or 3 major genes to enhance the field life of wheat cultivars.

5. THE CONCEPT OF DURABLE RESISTANCE

The problem of newly emerged races of pathogens has led to the adoption of alternative forms of resistance by the breeders that are more durable such as slow rusting or partial resistance [33]. It has been indicated that durable rust resistance is more likely to be of adult plant type rather than seedling type and is not associated with the genes conferring hypersensitive reaction [34]. Durable rust resistance is a mechanism conferring resistance to a cultivar for long period of time during its widespread cultivation in a favorable environment for a disease [35]. This type of resistance is mainly associated with the minor genes which are also known as slow rusting genes. The concept of slow rusting in wheat was proposed by reference [36], similar to partial resistance to late blight of potato put forth by reference [37]. Various workers have stressed the need to recognize and exploit longer-lasting resistance. Reference [35] defined durable resistance as a resistance source that remained effective after widespread deployment over a considerable period. A general concept of a durable resistance (a race non-specific) resistance source for a cereal rusts is as follow:

1) It may be controlled by more than a single gene;

2) It is more likely to operate at the adult-plant stage rather than at both the juvenile and adult stage;

3) It confers non-hypersensitive response to infection.

Example of durable resistance include resistance to stem rust transferred from tetraploid emmer to bread wheat Hope and H-44 [38], resistance to leaf rust in the South American wheat cultivar Frontana and related sources [15].

6. GENETIC BASIS OF DURABLE RESISTANCE

The durable resistance is based on additive effect of partial resistant minor genes, usually polygenic in nature and active in adult plant stage. Genetic studies conducted at CIMMYT, Mexico has shown that at least 10 - 12 different genes are involved in group of CIMMYT germplasm, and by accumulating 4 - 5 minor genes resistance level near to immunity can be achieved. However 2 - 3 genes in a line provide moderate level of resistance [39]. Most of these genes are undesignated only the genes *Lr*34/*Yr*18, *Lr*46/*Yr*29 and *Sr*2/*Yr*30 have been given names and designated to specific chromosomes. Each of these genes pairs are tightly linked or pleotropic. The varieties possessing minor gene based resistance show almost same level of resistance over space and time. For example Lyalpur 73 which was among major varieties of Pakistan in 1970's still show very good level of resistance in screening nurseries. Whereas, the varieties having major gene based race specific resistance did not have long life and collapsed usually after 4 - 5 years. Varieties having durable type of resistance show almost same level of reaction against different races and their resistance remained effective in different climatic conditions. The leaf rust and yellow rust reaction of some varieties having durable rust resistance are same at CIMMYT, El Batan, Mexico and Faisalabad, Pakistan. The variety Frontana being released about half century ago still have effective rust resistance almost every where. There are very rare examples that resistance based on major genes had been effective for a longer period of time. Reference [40] identified 6 independent loci, contributing to adult plant resistance (APR) or slow rusting contributing to two rusts in a population derived from cross of Avocet S and Pavon. The putative loci identified on chromosomes 1BL, 4BL and 6AL influenced resistance to both stripe and yellow rust. The loci on chromosome 3BS and 6BL had significant effect on stripe rust. The locus on the distal region of chromosome 1BL with highly significant effects had also detected in other mapping populations [34,41]. The distortion associated with chromosome 4B linkage map has also been observed in some other research reports [41]. Even Morocco and Avocet S have some genetic factors that contain some slow rusting resistance which results in significant delay in becoming completely susceptible [40]. The material having minor gene based resistance near to immunity for leaf and yellow rust was developed and distributed worldwide in 1990's by CIMMYT [33].

7. *Sr*2/*Yr*30 GENE

The gene, *Sr*2 was transferred to hexaploid wheat from tetraploid emmer wheat cultivar Yaroslav in 1920. It is present on chromosome 3BS and is also reported to be associated with *Lr*27 [42]. It is completely linked with pseudo black chaff (Pbc), which is used as morphological marker for identification of lines carrying this gene. The genotypes with Pbc show varying levels of stem rust infection. The maximum severity level of 60% - 70% has been noted as compared to 100% severity of susceptible check in disease screening nursaries in Kenya. When present alone it does not provide sufficient level of resistance but in combination with other genes desirable level of resistance can be achieved. Much information is not available about the interaction of *Sr*2 and other genes in *Sr*2 complex. The adequate resistance level can be achieved by accumulating 4 - 5 minor genes. *Sr*2 was detected in several highly resistant old, tall Kenyan cultivars like Kenya plume and semidwarf CIMMYT, cultivars Pavon F 76, Parula, Kingbird, Dollarbird etc. These cultivars show maximum disease severity of 10% - 15% with moderately resistant reactions. The gene *Sr*2 is tightly linked with *Yr*30 or has pleotropic effects [43]. A microsatellite (SSR) marker *gwm*533 is tightly linked and associated with the presence of this gene, which can be used to facilitate selection of this difficult to score gene [44].

8. *Lr*34/*Yr*18

Numerous genes conferring resistance to wheat rusts have been identified and used in wheat (*T. aestivum* L.) breeding. However, several of these genes have been rendered ineffective due to emergence of new virulent races. Cultivars with the rust resistance gene *Lr*34 such as Frontana had effective durable rust resistance to leaf rust (*P. triticina*). Although *Lr*34 has been used extensively in spring wheat grown in US, isolates of *P. tiriticina* with complete virulence to this gene had not been detected [45]. It has been found that soft red winter wheats having *Lr*34 in combination with seedling resistant *Lr*2a, *Lr*9, *Lr*26 were highly resistant while in combination with *Lr*10, *Lr*11, *Lr*18 were moderately to low resistant in USA [46].

Gene *Lr*34 first described by reference [8] has been shown to enhance leaf rust resistance in combination with other genes [47]. Another feature of *Lr*34 resistance is that it remained genetically inseparable from *Yr*18 gene, which confers APR. This gene co-segregates with leaf tip necrosis (*Ltn*1), powdery mildew resistance (*Pm*8), Barley yellow dwarf virus (*Bydv*1) genes [48,49]. These multipathogen resistance traits have made the *Lr*34/*Yr*18 locus one of the highly valuable regions for disease resistance in wheat [45]. If *Lr*34/*Yr*18 complex is present alone the disease level may go high but in combination with other genes it could give effective control [50]. At low temperature the resistance level conferred by plants with *Lr*34 is higher under growth chamber and green house condition. The gene seems to be effective under field conditions at average daily temp 0˚C - 20˚C and helps in reducing disease progress [24]. Reference [11] had indicated that environment has a significant influence on terminal disease reaction for leaf rust. Reference [16] showed that *Yr*18 may display inadequate resistance under some environmental conditions. It is present in many subcontinental varieties including some released in pre green revolution era. A marker associated with *csLV*34 locus on chromosome 7D was found associated with *Lr*34/*Yr*18 gene. Two predominant allelic size variants *csLV*34*a* and *csLV*34*b* were identified. A strong association was observed with the presence *Lr*34/*Yr*18 gene and csLV34b allele. However lines having *Lr*34/*Yr*18 gene and positive for csLv34a allele were rare. The lineage of this gene is tracked back to varieties Mentana and Ardito developed in Italy during early 1990's [45]. This gene has been cloned and was shown that *Lr*34/*Yr*18/*Pm*38/*Ltn*1 is the same gene [51].

9. *Lr*46/*Yr*29

A slow rusting gene identified in the cultivar Pavon and was found located on chromosome 1B by crossing with a monosomic series of adult plant leaf rust susceptible cultivar Lal Bahadur [52]. This is the 2nd named minor gene involved in slow rusting. The leaf rust resistance gene *Lr*46 and yellow rust resistance gene *Yr*29 are tightly linked or Pleotropic [53]. Its effect is similar to *Lr*34/*Yr*18 as it does not provide complete immunity to plants. Infected adult plants carrying *Lr*46 have longer latency period as compared to control without this gene [54]. The plants with this gene also show higher rate of fungal colonies abortion with out any chlorotic or necrotic effects and also decrease the colony size. The resistance conferred by this gene is not of hypersensitive. Reference [41] determined that the microsatellite locus

*Xwmc*44 is located 5.6-cM proximal to the putative QTL for *Lr*46. Leaf tip necrosis (Ltn) has been reported to be highly correlated with the presence of *Lr*46/*Yr*29 [55]. Efforts are underway to clone this gene (www.ars.usda.gov).

10. BREEDING FOR DURABLE RUST RESISTANCE

Accumulating minor genes for attaining desired level of resistance in a variety is a challenging task [56] as it requires identification of parents with minor genes, crossing them in specific schemes following back cross or top cross approach, maintaining desirable population size and selection of desirable genotypes from segregating populations. The crossing schemes and selections strategies used for breeding major genes based resistance are not suitable for breeding minor gene resistance.

The modified pedigree method used for breeding major gene based resistance can not give any progress for minor gene based resistance. Reference [52] compared different crossing and selection schemes for working out their efficiency in terms of genetic gains and cost efficiency. The influence of type of cross and selection scheme was minimal on main grain yield. They found that selection of parents was the most important feature in breeding for achieving desirable results. They also reported that mean rust severity of top cross progenies was less as compared to simple cross because two parents contributed resistance factors to the top cross progenies. Non selected bulk method was found to be least effective and selected bulk method as the most attractive schemes in terms of genetic gain and cost efficiency. An example of breeding for minor gene based resistance is the development of wheat stock resistant to leaf and yellow rust at CIMMYT.

Since early days of breeding for minor genes, plants and lines with infection intensity of 20% - 30% and compatible infection type were targeted. This led to the development of wheat varieties Nacozari F 76, Pavon F 76 and several others [56] which were released not only in Mexico but also in Ethiopia, Bangladesh, Pakistan and other countries. Pavon was released in 16 countries with different names (**Table 2**). This material provided the foundation for breeding for minor gene resistance.

In Pakistan the varieties Uqab 2000 (CROW'S'/NAC/BOW'S'), Bhakkar-02 (P-102/PIMA//F3.71/TRM/3/PVN) and Seher-06 (CHIL/2*STAR/BOW/ CROW/BUC/PVN/3/VEE#10) have this type of resistance. Bhakkar-02 has dominated the mega wheat cultivar Inqilab91 since 2005 after Inqilab 91 was hit by yellow rust epidemic and Seher-06 is gaining popularity now, due to its higher yield and better resistance to leaf and yellow rusts. The variety,

Table 2. Derivatives of Pavon (VCM/CNO/7C/3/KAL/BB) released in different part of the world.

S. No	Country	Variety	Year	Pedigree
1	Algeria	Citra 78	1978	CM8399-D-4M-3Y-1M-1Y-0Y-0DZA
2	Algeria	Chellif 78	1978	CM8399-D-4M-3Y-0M-0DZA
3	Bangladesh	Pavon 76-Bgd	1979	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0BGD
4	Bolivia	Pilanchu 80	1980	CM8399-D-4M-3Y-1M-0M-1-14Y-0BOL
5	Bolivia	Totora Ibta 80	1980	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0BOL.
6	Chile	Victoria	1981	CM8399-D-4M-3Y-0M-0CHL
7	Chile	Marib 1	1983	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0YMD
8	Chile	Onda Inia	1982	CM8399-D-4M-3Y-1M-1Y-0M-0CHL
9	Egypt	Giza 162	1988	CM8399-0 EGY
10	Ethiopia	Pavon 76-Eth	1982	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0ETA
11	Mexico	Pavon F 76	1976	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0MEX.
12	Morocco	Baraka	1988	CM8399-0MAR
13	Nigeria	Samwhit 6	1990	CM8399-0NER
14	Pakistan	Pavon Pak	1978	CM8399-D-4M-3Y-1M-1Y-1M-0Y-0PAK
15	Peru	Elgavilon	1982	CM8399-D-4M-3Y-1M-1Y-0Y-0PER
16	Tanzania	Azimio 87	1987	CM8399-D-4M-3Y-1M-1Y

Uqab 2000 proved the best option for the rain fed northern Pakistan after severe epidemic of yellow rust in 2005.

An example of breeding for durable rust resistance out side CIMMYT is the wheat breeding program of Ayub Agricultural Research Institute, Faisalabad-Pakistan. The germplasm of wheat (about 1200 accessions) was screened under artificial inoculation with mixture of races and the parents having partial resistance for leaf/yellow rust were selected [57]. This germplasm was crossed to pyramid genes for high yield and rust resistance. The main focus was accumulation of minor genes for rust resistance because this type of resistance mechanism is considered more durable and is effective for many races rather than single race [58]. The parents selected were used to construct back crosses, top crosses and double crosses. Mostly selected bulk method was used as described by reference [39] to advance the filial generations to conserve maximum genetic diversity. In F_2 generation, 2500 - 3000 plants were raised. Heads were taken from plants having desirable plant type and were bulked to raise F_3 generations. From F_3 - F_6 generations, plants were selected on the basis of good plant type and rust intensity. The heads were taken from the plants having rust intensity ranging 0% - 30% preferably with R/MR/MS type of reaction.

Various lines were selected from this material and tested for yield and disease reaction. Two varieties Shafaq-06 and Lasani-08 having durable type of resistance were approved for general cultivation in the Punjab-Pakistan [59,60]. These varieties are high yielding and possess durable resistance to leaf and yellow rust. Lasani-08 was also found resistance to stem rust (Ug 99) in the year 2007 at Kenya.

11. MARKER-ASSISTED SELECTION (MAS) AND RESISTANCE GENE ISOLATION AS TOOL FOR IMPROVING THE RESISTANCE TO LEAF AND STEM RUST RESISTANCE IN WHEAT

Pyramiding of several genes into one cultivar can be an effective strategy to use resistance genes to enhance durability of wheat resistance to leaf and stem rust [3]. Durable resistance may be achieved by combination of several genes encoding partial resistance. Gene pyramiding through conventional methods is difficult and time-consuming because it requires simultaneous tests of the same wheat breeding materials with several different rust races before a selection is made. Usually, it is not feasible for a regular breeding program to maintain all necessary rust races needed for this type of work. Therefore, MAS is a powerful alternative to facilitate new gene deployment and gene pyramiding for quick release of rust-resistant cultivars. Molecular markers such as STS or SCAR and CAPS are available for leaf rust resistance genes *Lr*1, *Lr*9, *Lr*10, *Lr*19, *Lr*21, *Lr*24, *Lr*25, *Lr*28, *Lr*29, *Lr*34, *Lr*35, *Lr*37, *Lr*39, *Lr*47 and *Lr*51. Enzymatic marker (endopeptidase Ep-D1c) for *Lr*19 has also been developed [61]. Microsatellite (SSR) and AFLP markers for some *Lr* genes such as *Lr*3bg, *Lr*18, *Lr*40, *Lr*46 and *Lr*50 have been developed by reference [62,63]. Molecular markers are available also for stem resistance genes such as *Sr*2, *Sr*9a, *Sr*22, *Sr*24, *Sr*26, *Sr*31, *Sr*36 and *Sr*39. Some of the markers have been used in MAS, but markers for some of the genes are not diagnostic for the genes and must be improved and markers for other genes are not available. At the present time, the research of stem rust in wheat has focused on identifying more resistance genes to control Ug99. According to the Farm and Ranch Guide report, currently 50% of winter wheat and 70% to 80% of spring wheat used in the USA are susceptible to Ug99. Moreover, 75% - 80% of the breeding materials are susceptible to Ug99 and most stem rust resistance genes deployed in breeding programs have been overcome by this new fungus (http://www.farmandranchguide.com/articles/2008/03/13/ag_news/ production_news/pro10.txt).

Microsatellite marker closely linked to resistance gene Sr40 has been also obtained [64,65]. Date three genes for leaf rust resistance in wheat *Lr*1, *Lr*10 and *Lr*21 [66] have been isolated cloned and sequenced. They all have sequences that encode nucleotide binding site (NBS)-leucine-rich repeat (LRR) regions, which are characteristic of disease resistance genes in plants. Molecular description of these genes in wheat provides a unique biological system to study the molecular mechanisms of wheat-pathogen interaction and transduction as well as the resistance gene function, evolution and diversity. This will allow further manipulation of wheat resistance genes to increase the resistance durability by genetic transformation of wheat.

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