Meta-QTLs for multiple disease resistance involving three rusts in common wheat (*Triticum aestivum* L.)

Neeraj Pal1 · Irfat Jan2 · Dinesh Kumar Saini3 · Kuldeep Kumar2 · Anuj Kumar2 · P. K. Sharma2 · Sundip Kumar1 · H. S. Balyan2 · P. K. Gupta2,4

Received: 25 January 2022 / Accepted: 28 April 2022 / Published online: 14 June 2022
© The Author(s), under exclusive licence to Springer-Verlag GmbH Germany, part of Springer Nature 2022

Abstract

**Key message** In wheat, multiple disease resistance meta-QTLs (MDR-MQTLs) and underlying candidate genes for the three rusts were identified which may prove useful for development of resistant cultivars.

Abstract Rust diseases in wheat are a major threat to global food security. Therefore, development of multiple disease-resistant cultivars (resistant to all three rusts) is a major goal in all wheat breeding programs worldwide. In the present study, meta-QTLs and candidate genes for multiple disease resistance (MDR) involving all three rusts were identified using 152 individual QTL mapping studies for resistance to leaf rust (LR), stem rust (SR), and yellow rust (YR). From these 152 studies, a total of 1,146 QTLs for resistance to three rusts were retrieved, which included 368 QTLs for LR, 291 QTLs for SR, and 487 QTLs for YR. Of these 1,146 QTLs, only 718 QTLs could be projected onto the consensus map saturated with 2, 34,619 markers. Meta-analysis of the projected QTLs resulted in the identification of 86 MQTLs, which included 71 MDR-MQTLs. Ten of these MDR-MQTLs were referred to as the ‘Breeders’ MQTLs’. Seventy-eight of the 86 MQTLs could also be anchored to the physical map of the wheat genome, and 54 MQTLs were validated by marker-trait associations identified during earlier genome-wide association studies. Twenty MQTLs (including 17 MDR-MQTLs) identified in the present study were co-localized with 44 known R genes. In silico expression analysis allowed identification of several differentially expressed candidate genes (DECGs) encoding proteins carrying different domains including the following: NBS-LRR, WRKY domains, F-box domains, sugar transporters, transferases, etc. The introgression of these MDR loci into high-yielding cultivars should prove useful for developing high yielding cultivars with resistance to all the three rusts.

Communicated by Thomas Miedaner.

Neeraj Pal and Irfat Jan have contributed equally to this work.

* P. K. Gupta
pkgupta36@gmail.com

1 Department of Molecular Biology and Genetic Engineering, G. B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand 263145, India
2 Department of Genetics and Plant Breeding, Ch. Charan Singh University, Meerut 250004, India
3 Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana, Punjab 141004, India
4 Murdoch’s Centre for Crop & Food Innovation, Murdoch University, Murdoch, WA 6150, Perth, Australia

Introduction

Wheat production suffers major losses due to three rusts, namely leaf rust (LR, caused by *Puccinia triticina* f. sp. *tritici*), stem rust (SR, caused by *P. graminis* f. sp. *tritici*), and yellow rust (YR, caused by *P. striiformis* f. *sp tritici*) (Joshi et al. 1985; Rana et al. 2021). According to available estimates, LR causes yield losses of up to 50% in favorable conditions (Knott 1989; Huerta-Espino et al. 2011), SR causes yield losses of up to 100% during epidemic outbreaks (Soko et al. 2018), and YR causes yield losses ranging from 10 to 70%. The extent of yield losses in these estimates sometimes also depends upon the cultivar used, time of infection, rate of disease development, and duration of the disease (Chen 2005; Afzal et al. 2007). Chemical control and development of resistant cultivars are the two major options for managing the yield losses as above. Since the pathogens develop resistance against fungicides rather quickly (Oliver 2014), the development of resistant cultivars is the preferred approach to overcome the losses due to rust infections. This requires adequate knowledge of the genetics of resistance.

A large number of studies have already been conducted to identify genes for resistance to individual diseases (Li et al. 2020). As a result, R genes or gene systems have become available for resistance against different diseases. These
R genes can be race-specific or race-non-specific, the latter providing broad-spectrum resistance (Kou and Wang 2010; Kaur et al. 2021). Currently, a large number of R genes in wheat are known for each of the three rusts, which include ~80 genes for LR, ~70 genes for SR and ~83 genes for YR (McIntosh et al. 2016; Pradhan et al. 2020). These genes have regularly been used for the improvement of disease resistance, although only a fraction of these genes has been utilized in developing resistant cultivars. The development of resistant cultivars generally involves transfer of one or more genes for a specific disease, although efforts have also been made to develop resistance for multiple diseases in a breeding programme. This has generally been achieved through pyramiding of genes (Gupta et al. 2021; Sharma et al. 2021; Rana et al. 2021; Pal et al. 2022). The method of pyramiding genes through conventional breeding is, however, quite demanding and requires a long time to achieve the desired level of resistance. In order to overcome this limitation, the concept of multiple disease resistance (MDR) was put forward initially for legumes (Nene 1988), followed by several other reports in different crops including wheat (Singh et al. 2012; Zwart et al. 2010; Jighly et al. 2016; Mago et al. 2011). MDR differs from broad-spectrum resistance and APR, both of which generally provide resistance against most prevalent races for a particular pathogen, and not resistance against several pathogens (Wiesner-Hanks and Nelson 2016).

MDR has been described as "the holy grail" for several crops including barley (Paterson 2014), and wheat (Pooja et al. 2014). Inheritance of MDR has also been studied (Ali et al. 2013; Pooja et al. 2014; Schweizer and Stein 2011; Wiesner-Hanks and Nelson 2016); in these inheritance studies, the occurrence of MDR has been inferred from a high level of positive correlations observed among resistance to individual diseases within the same crop (Randhawa et al. 2018). This MDR has been attributed to either the tightly linked clusters of R genes or pleiotropy. Some of the examples of tightly linked genes in wheat for resistance against several diseases involving three rusts include the following: (i) *Lr34/Yr18/Sr57/Pm38/Ltn1* (7DS; Krattinger et al. 2009), (ii) *Lr46/Yr29/Sr58/Pm39/Ltn2* (1BL, Singh et al. 2013), (iii) *Sr2/Lr27/Yr30/Pbc1* (3BS; Mago et al. 2011), (iv) *Lr67/Yr46/Sr55/Pm46/Ltn3* (4DL; Herrera-Foessel et al. 2014).

MDR can also be regarded as an exceptional form of broad-spectrum resistance and is thus potentially difficult to overcome (Wiesner-Hanks and Nelson 2016). Two widely known examples of MDR genes include recessive gene *mlo* in barley and dominant gene *Lr34* in wheat, the latter providing resistance to several diseases in wheat (Krattinger et al 2009; Lagudah et al 2009). *Lr34* has been effective and has been widely used for more than 100 years. The complex locus *Lr34/Yr18/Pm38* is also a MDR locus, but when it was cloned in 2009, MDR was found to be conferred by a single gene encoding an ATP-binding cassette (ABC) transporter (Krattinger et al 2009). Although neither the substrate of the ABC transporter nor the mechanism by which it provides resistance is known, it is speculated to have a role in transporting or sequestering xenobiotic compounds.

MDR has also been identified in naturally occurring wheat cultivars. For instance, long back in the history, wheat cultivars Hope and H-44 were both shown to carry resistance to not only LR and SR, but also for loose smut and covered smut (Ausemus 1943). Multi-trait (MT) analysis for quantitative disease resistance (QDR) involving more than one correlated diseases also allowed identification of QTLs conferring resistance to multiple diseases, suggesting that there may be complex loci, which control resistance against more than one diseases (Singh et al. 2012). In some recent reports, mainly involving genome-wide association studies (GWAS), MDR has also been reported in 5–10% of the naturally occurring wheat genotypes, including some synthetic wheats (Friesen et al. 2008; Gurung et al. 2009; Miedaner et al. 2020; Kumar et al. 2020b; Saini et al. 2022a). The details of these reports are summarized in Table S1. The source of MDR identified in wheat genotypes used for several GWA studies may not be known, but we assume that pyramiding of genes through conventional breeding or occurrence of tight linkage or pleiotropy should be responsible for MDR in these cultivars. In recent years, the pyramiding of genes for resistance against more than one diseases has also been facilitated through markers assisted selection (MAS). One good example is the pyramiding of genes for thousand-grain weight (TGW) over three rust resistance genes (*Yr15* from one source and *Lr57-Yr40* from another source) into two popular Indian cultivars, namely PBW343 and PBW550 (Kaur et al. 2020).

The idea of MDR also prompted studies involving identification of meta-QTLs using known QTLs for each of several individual diseases. This resulted in identification of MDR-MQTLs in barley (Schweizer and Stein 2011), maize (Ali et al. 2013), rice (Kumar and Nadarajah 2020), and wheat (Saini et al. 2022a). In the present study, meta-QTL analysis was conducted for identification of MDR-MQTLs providing resistance to three rusts, namely LR, SR, and YR in wheat; for this purpose, QTLs reported for resistance against all the three rusts were utilized. MDR-MQTLs thus identified were also compared with MTAs reported earlier using GWAS and with the available results of transcriptomics undertaken to uncover potential genomic regions and key candidate genes (CGs) that influence MDR in wheat. We hope that the MDR MQTLs identified during the present study should prove useful for the transfer and clustering of MQTLs and CGs for achieving MDR in wheat breeding programs.
Materials and methods

Collection of information on QTLs associated with resistance against LR, SR, and YR

Using PubMed and Google Scholar, an extensive search was made for QTLs already reported to be associated with resistance to LR, SR, and YR (till August 2021). The information thus obtained was further supplemented by the wheat QTL database (WheatQTLdb; www.wheatqtldb.net; Singh et al. 2021). Following information about individual QTLs was collected and compiled from each such study: (i) type and size of the mapping population, (ii) flanking markers and their genetic positions on the map, (iii) peak positions, (iv) phenotypic variation explained (PVE) or R² value, and (v) logarithm of the odds (LOD) score for each QTL. In cases, where no information was available on the peak position of QTL, the mid-position of the two flanking markers was taken as the peak. Also, when the confidence interval (CI) for the QTL was missing, the CI (95%) was estimated using the following population-specific equations developed through different simulations:

(i) F₂ and BC populations (Darvasi and Soller 1997):

\[
\text{CI (95\%)} = \frac{530}{(R^2 \times N)}
\]

(ii) For DH populations (Visscher and Goddard 2004):

\[
\text{CI (95\%)} = \frac{287}{(R^2 \times N)}
\]

(iii) For RIL populations (Guo et al. 2006):

\[
\text{CI (95\%)} = \frac{163}{(R^2 \times N)}
\]

From each study, the following two types of input data text files were prepared using the instructions available in the BioMercator v3 manual (Sosnowski et al. 2012): (i) genetic map file and (ii) QTL information file. The map file thus generated for each study included information on the type and size of the population, mapping function, map units, and positions of different markers on linkage groups. QTL information file included information on QTL names, associated diseases, chromosome names, PVE values of individual QTLs, genetic positions, etc. Studies that lacked essential data (e.g., PVE or R²; marker positions) were excluded from the analysis.

Construction of the consensus map and QTL projection

The consensus genetic map was constructed using R package “LPMmerge” (Endelman and Plomion 2014) utilizing following high-quality genetic maps: (i) ‘Wheat Consensus SSR 2004’ map with 1235 markers (Somers et al. 2004), (ii) ‘Wheat Composite 2004’ map with 4403 markers, available at the GrainGenes database (http://wheat.pw.usda.gov), (iii) ‘Integrated 2013’ durum wheat map with 3669 markers (Marone et al. 2013), and (iv) four SNP maps developed using following different SNP arrays: ‘Illumina 9 K iSelect Beadchip Array’ (with 7,504 SNPs) (Cavanagh et al. 2013), the ‘AxiomR, Wheat 660 K SNP array’ (with 1,19,566 SNPs) (Cui et al. 2017), ‘Illumina iSelect 90 K SNP Array’ (46,977 SNPs) (Wang et al. 2014), and ‘Wheat 55 K SNP array’ (with 56,505 SNPs) (Winfield et al. 2016). Additional markers flanking each QTL reported in individual studies for LR, SR, YR, and resistance (R) genes were also included on the consensus genetic map.

QTLs were projected onto the above consensus map using the projection tool (QTLProj) available in Biomercator v4.2 (Chardon et al. 2004); QTLProj uses a dynamic algorithm to determine the best context for projection. An optimal context consists of a pair of common markers that flank the QTL in the original map and for which the distance is consistent between the original map and the consensus map. The behavior of the algorithm to find such a configuration is controlled by the lower values of the ratio of the distances between flanking markers and of the p-value obtained by testing the homogeneity of these distances in the original map and the consensus maps (Veyrieras et al. 2007).

Identification of MQTLs

Meta-QTL analysis was performed using the position of each input QTL and the variance of this position, assessed through CI values; the analysis was based on Biomercator v4.2 (Arcade et al. 2004; Sosnowski et al. 2012). Two different approaches were used, one involving cases, where the number of QTLs on an individual chromosome was ≤ 10, (Goffinet and Gerber 2000), and the other including cases, where this number was > 10 (Veyrieras et al. 2007). The model with the lowest Akaike information criterion (AIC) value was chosen as the best fit in the first approach. In the second approach, the best model was chosen from among the following four models: AIC model, corrected AIC (AICc and AIC3) model, Bayesian Information Criterion (BIC) model, and Average Weight of Evidence (AWE) model.

MQTLs were named based on their genetic positions; for instance, MQTLs mapped on chromosome 1A were designated as MQTL1A.1, MQTL1A.2, and so on. The PVE value and LOD score of a MQTL were calculated as the mean of the PVE values and LOD scores of the QTLs, on which the MQTL is based. The nucleotide sequences of flanking markers of each MQTL were BLASTed against the wheat reference genome sequence to find out their physical positions in the genome. Physical positions of GBS-SNPs were obtained using the JBrowse wheat genome browser (https://wheat-urgi.versailles.inra.fr/Tools/Jbrowse).

Comparison of MQTLs with GWAS-based MTAs

MQTLs identified in the present study were compared with MTAs reported from 23 independent GWA studies.
(including 4 GWA studies on LR, 6 on SR, 7 on YR, 6 studies including at least two different rusts). These GWA studies utilized populations of durum wheat, hexaploid wheat (e.g., spring, and winter wheat) and synthetic hexaploid wheats (SHW) with population sizes ranging from 100 (Leonova et al. 2020) to 23,346 (Juliana et al. 2020), phenotyped across the 17 different countries. The details of population size, associated diseases, genotyping platform/number of SNP markers used, and MTAs detected in these studies are available in Table 1. Physical positions of each significant and stable SNP associated with the trait were obtained from the respective studies or JBrowse-WHEAT URGI database (https://urgi.versailles.inra.fr/jbrowsewgsr/) and CerealsDB (https://www.cerealsdb.uk.net/cerealgenomics/CerealsDB/indexNEW.php). Subsequently, physical positions of these MTAs were compared with physical coordinates of the MQTLs; MQTL co-localizing each with at least one MTA was considered as GWAS-validated/verified MQTL.

**Table 1** Summary of genome-wide association studies in wheat on LR, SR, and YR used in the present study

| Type of wheat (Panel size) | Genotyping platform/Number of MTA's | Country | References |
|----------------------------|------------------------------------|---------|------------|
| **Leaf rust, stem rust and yellow rust** | | | |
| Common wheat cultivars from Kazakhstan and Europe (215) | 20 K (11,510 SNPs) | 44 Kazakhstan and Russia | Genievskaya et al. (2020) |
| Spring wheat (483) | 35 K (14,650 SNPs) | 438 India | Kumar et al. (2020a) |
| Spring wheat (148) | 9 K (5688 SNPs) | 79 Australia | Kankwatsa et al. (2017) |
| **Leaf rust and yellow rust** | | | |
| Wheat accessions (268) | 90 K (12,931 SNPs) | 560 China | Zhang et al. (2021) |
| Winter durum wheat (328) | DArT (12,550) | 7 Germany, Austria, and Hungary | Miedaner et al. (2019) |
| **Yellow rust and stem rust** | | | |
| European winter wheat diversity panel (158) | DArTs, SNP (21,543) | 61 Germany | Miedaner et al. (2020) |
| **Leaf rust** | | | |
| Durum, tetraploid, and hexaploid winter wheat accessions (385) | 90 K (9570 SNPs) | 133 Canada | Fatima et al. (2020) |
| Spring wheat (100) | 15 K (NA) | 16 Russia | Leonova et al. (2020) |
| Spring wheat and soft red winter wheat accessions (331) | 9 K (5025 SNPs) | 50 Georgia | Sapkota et al. (2019) |
| Leaf rust association mapping panel (381) | 90 K (18,925 SNPs) | 82 Minnesota (USA) | Gao et al. (2016) |
| **Stem rust** | | | |
| ICARDA spring wheat (245) | 15 K (9523 SNP) | 44 Ethiopia | Shewabez et al. (2021) |
| Spring wheat panel (250) | GBS (9042 GBS) | 66 Ethiopia and Germany | Edae et al. (2020) |
| Spring durum wheats (283) | GBS (26,439 SNPs) | 126 Ethiopia, Kenya | Megerssaa et al. (2020) |
| Iranian bread wheats (282) | GBS (8,959) | 8 Iran | Saremizad et al. (2020) |
| Durum wheats (183) | SSR, DArT, STS (953) | 8 Ethiopia | Letta et al. (2013) |
| Winter wheats from 1st and 3rd WWSRRNs (232) | DArT (4721 DArT) | 7 Kenya | Yu et al. (2012) |
| **Yellow rust** | | | |
| Durum wheat landraces and cultivars (300) | 35 K (7093 SNPs) | 35 Ethiopia | Alemu et al. (2021) |
| Spring wheat genotypes (465) | GBS (765 SNPs) | 25 Pakistan, USA | Habib et al. (2020) |
| Chinese wheat cultivars (240) | 90 K (NA) | 21 Wuhan (China) | Jia et al. (2020) |
| Advanced wheat breeding lines (CIMMYT) (23,346) | GBS (78,662 SNPs) | 31 India, Kenya and Mexico | Juliana et al. (2020) |
| CIMMYT pre-breeding wheat lines (419) | DArTseq (22,415) | 14 Mexico | Ledesma-Ramírez et al. (2019) |
| Spring wheat accessions (1000) | 9 K (4585 SNPs) | 40 Washington state (USA) | Maccarelli et al. (2015) |
| Synthetic wheat accessions and bread wheat cultivars (192) | 9 K (2590 SNPs) | 31 Ethiopia | Zegeye et al. (2014) |
Association of MQTLs with known R genes

The association of MQTLs with known R genes for the three rusts was also examined. For this purpose, sequences of R genes or their associated markers were BLASTed against the wheat reference genome to obtain the physical positions of the genes. The physical positions of the genes were then compared with the physical coordinates of the MQTLs to ascertain the co-localization of MQTLs with known R genes.

Identification of candidate genes (CGs) in MQTL regions and their GO analysis

The BioMart tool of Ensembl Plants database (https://plants.ensembl.org/biomart/martview) was used to identify CGs within 2 Mb regions flanking 1 Mb on either side of the peaks of the individual MQTLs. In some cases, peaks of individual MQTLs had to be worked out before the search for CGs using methods reported earlier (Saini et al. 2022c). The InterPro database (https://www.ebi.ac.uk/interpro/) was used to extract the physical coordinates, function descriptions, and gene ontology (GO) terms of the identified gene models.

In silico expression analysis of the candidate genes

In silico expression analysis of the above CGs was conducted using expVIP (http://www.wheat-expression.com/; Ramirez-Gonzalez et al. 2018) for stripe rust and GENEVESTIGATOR database (https://genevestigator.com/; Hruz et al. 2008) for leaf and stem rusts. Unfortunately, expression data for CGs were not available for all the three rusts in the same database. In the GENEVESTIGATOR database, 11 studies were available and only 4 studies carried the expression data of our putative CGs (Cloutier et al. 2008; Rutter et al. 2017; Salcedo et al. 2017; Yadav et al. 2016). The expression data for YR resistance belonged to two different studies (Zhang et al. 2014; Dobon et al. 2016). The data on expression were available as log2 transformed tpm (transcripts per million) values. Only CGs showing FC ≥ 2 (upregulation, twofold or more) or FC ≤ −2 (downregulation, twofold or more) were accepted as showing differential expression in the form of fold changes estimated by comparing tpm values under stress vs. control. The representative heatmap of the DECGs was generated using an online tool ClustVis (https://biit.cs.ut.ee/clustvis/).

Homoeologous relationship

Homoeologous relationships of MQTLs were also examined, using corresponding physical positions on the known homoeologous chromosomes. For this purpose, genes were identified from the complete MQTL region, and these genes were subjected to BLAST analysis against the wheat genome database available at EnsemblPlants to identify the corresponding homoeologs on different wheat chromosomes. Homoeologs were extracted with their physical positions from the database, which were then compared with the physical coordinates of the MQTLs. MQTLs located on homoeologous chromosomes having similar genes were accepted as homoeologous MQTLs.

Results

QTLs associated with LR, SR, and YR

Information of 1,146 wheat QTLs (368 for LR, 291 for SR, and 487 for YR) from 152 interval mapping studies was collected (Table S2). These 152 studies included 31 studies for LR, 24 studies for SR, 66 studies for YR, and 31 studies each associated with at least two different rusts (Table S2, Fig. 1a). A total of 160 mapping populations were utilized in these 152 studies, which included 117 sets of RILs, 31 sets of DH lines, and 12 F3/backcross populations (some studies utilized more than one population). The population size in different studies ranged from 58 (Zhang et al. 2016) to 1020 (Agenbag et al. 2014), and the number of QTLs in individual studies ranged from one each in several individual studies (e.g., Kolmer et al. 2011; Wang et al. 2015; Zhang et al. 2016; Zwart et al. 2008) to a maximum of 59 QTLs in one study (Bajgain et al. 2016). The distribution of QTLs for three individual rusts and among 21 chromosomes along with the distribution of QTLs according to PVE and LOD score is presented in Fig. 1.

Consensus map for QTL projection

The three maps (other than the SNP maps) shared 566 markers, whereas ‘Wheat Composite 2004’ had 3531 unique markers, ‘Wheat Consensus SSR 2004’ had 81 unique markers, and ‘Integrated 2013’ durum wheat map had 2351 unique markers. The number of unique and shared (or common) markers among these maps is shown in a Venn diagram (Fig. 2). Markers showing ordinal conflicts were removed while constructing the consensus map using LPmerge package of R programming. The consensus map developed for the present study included 234,619 loci, which included loci for several different types of markers (SNPs, DArT, SSR, AFLP, RAPD, STS, EST-SSR, SRAP, ISSR, KASP) as well as gene loci (Vrn, Ppd, Rht, Glu, etc.) (Table S3). The length of the consensus map was 7637.09 cM (Fig. S1), giving a density of 30.72 loci/cM (Table S4). The marker densities for individual chromosomes are presented in Fig. S1. The marker density for
individual chromosomes ranged from 7.78 markers/cM for 4D to 71.62 markers/cM for 1A (Table S4). Sub-genome ‘A’ had a genetic length of 2565.38 cM with 35.8 markers/cM, sub-genome ‘B’ had a length of 2938.05 cM with 36.35 markers/cM, and sub-genome ‘D’ had a length of 2133.66 cM with 16.85 markers/cM.

For QTL projection, only 718 QTLs, which had flanking markers available on the consensus map, were used. These projected QTL included 175 QTLs for LR, 222 QTLs for SR, and 321 QTLs for YR. The projected QTLs on three sub-genomes were 210 for A sub-genome, 367 for B sub-genome and 141 for D sub-genome. Similarly, the number of projected QTLs for individual chromosomes ranged from 9 (each on 4D and 6D) to 93 (2B) with an average of 34 QTLs per chromosome. Mean CIs in original (or total collected QTLs) and projected QTLs (part of the total collected QTLs actually projected on to the consensus map) also differed for LR, SR, and YR (Fig. 3). For LR, 39.67% of the original and 24.45% projected QTLs had CIs greater than 10 cM. Similarly for SR, 20.62% of the original and 18.55% of the projected QTLs possessed CI values greater than 10 cM. Lastly, for YR, 32.85% of the original and 21.97% of the projected QTLs had CIs greater than 10 cM.

**MQTLs and their salient features**

The MQTL analysis gave 86 MQTLs, which involved only 596 of the 718 QTLs that were projected (Fig. 4, Table 2, 36.35 markers/cM, and sub-genome 'D' had a length of 2133.66 cM with 16.85 markers/cM.

For QTL projection, only 718 QTLs, which had flanking markers available on the consensus map, were used. These projected QTL included 175 QTLs for LR, 222 QTLs for SR, and 321 QTLs for YR. The projected QTLs on three sub-genomes were 210 for A sub-genome, 367 for B sub-genome and 141 for D sub-genome. Similarly, the number of projected QTLs for individual chromosomes ranged from 9 (each on 4D and 6D) to 93 (2B) with an average of 34 QTLs per chromosome. Mean CIs in original (or total collected QTLs) and projected QTLs (part of the total collected QTLs actually projected on to the consensus map) also differed for LR, SR, and YR (Fig. 3). For LR, 39.67% of the original and 24.45% projected QTLs had CIs greater than 10 cM. Similarly for SR, 20.62% of the original and 18.55% of the projected QTLs possessed CI values greater than 10 cM. Lastly, for YR, 32.85% of the original and 21.97% of the projected QTLs had CIs greater than 10 cM.

**MQTLs and their salient features**

The MQTL analysis gave 86 MQTLs, which involved only 596 of the 718 QTLs that were projected (Fig. 4, Table 2,
The remaining 122 projected QTLs included (i) 110 QTLs, for which the predicted QTL peaks were not included within any MQTL, and (ii) 12 singletons, each with solitary QTL in a predicted MQTL (Table S6). The 86 MQTLs were distributed on 20 wheat chromosomes (except 6D) (Fig. 4); the number of QTLs per chromosome ranged from a minimum of only two MQTLs each on 3D, 6B, 7A, and 7D to a maximum of 9 MQTLs on 6A (Fig. 5a). The number of MQTLs did not depend on the number of QTLs on individual chromosomes utilized for MQTL analysis; for instance, chromosomes 3B, 6B, and 7D each carried many more QTLs but relatively fewer MQTLs (Table S5). Only 71 of the 86 MQTLs could be classified as MDR-MQTLs, which included 28 MQTLs for all the 3 rusts, and 43 for two rusts each (8 for LR and SR; 14 for LR and YR; 21 for SR and YR) (Fig. 5b). The remaining 15 MQTLs each provided resistance for only one rust (1 for LR; 6 for SR and 8 for YR) (Table 2, Table S5). The number of QTLs per MQTL ranged from ≤ 5 in each of 52 MQTLs to ≥ 20 QTLs in each of three MQTLs (MQTL2B.3, MQTL3B.1, and MQTL7D.1) (Fig. 5c); 34 MQTLs that were each based on QTLs from four or more studies should be more stable across environments.

The CI of the MQTLs ranged from 0.04 to 14.3 cM with a mean of 1.58 cM, while that of projected QTLs ranged from 0.008 to 83.5 cM with a mean of 10.57 cM. There were significant differences in average CIs of MQTLs that mapped on different chromosomes, the reduction in mean CI ranged from 2.46-fold for MQTLs located from ≤ 5 in each of 52 MQTLs to ≥ 20 QTLs in each of three MQTLs (MQTL2B.3, MQTL3B.1, and MQTL7D.1) (Fig. 5c); 34 MQTLs that were each based on QTLs from four or more studies should be more stable across environments.

The CI of the MQTLs ranged from 0.04 to 14.3 cM with a mean of 1.58 cM, while that of projected QTLs ranged from 0.008 to 83.5 cM with a mean of 10.57 cM. There were significant differences in average CIs of MQTLs that mapped on different chromosomes, the reduction in mean CI ranged from 2.46-fold for MQTLs located on 5A to 21.6-fold for MQTLs located on 6B; while the mean reduction in the length of CI of all MQTLs was observed 6.69 fold (Fig. 5d). These lengths of CI in cM correspond to a physical distance, which ranged from 19 Kb (MQTL6A.1) to 604.3 Mb (MQTL2D.1) with a mean of 51.3 Mb in 78 MQTLs (only 78 of 86 were physically anchored); CI in 47 of these MQTLs each covered a physical distance of < 20 Mb. The LOD score of individual MQTLs ranged from 3.09 to 28.05, while the PVE ranged from 4.74 to 51.02%.

Ten MQTLs were selected to be the most important for breeders and were, therefore, named breeders’ MQTLs. Following criteria were used for the identification of breeders MQTLs: (i) MQTLs involving QTLs for all the three rusts, (ii) CI < 2 cM, (iii) average PVE > 15%, (iv) average LOD > 8, and, (v) involvement of at least 5 QTLs within the MQTL. These ten MQTLs included the following: MQTL1B.1, 2A.3, 2A.5, 2B.1, 2B.2, 2B.3, 3B.1, 4A.1, 6A.1, and 7D.2 (Table 3).

MQTLs overlapping GWAS-MTAs

In order to identify MQTLs with a higher level of confidence, the above 78 MQTLs were compared with 1,926 MTAs for the three rusts, reported in 23 GWAS; this resulted in the identification of 54 MQTLs overlapping 497 MTAs (Table S7); of these 54 MQTLs, 22 MQTLs each overlapped MTAs for all the three rusts, while 17 MQTLs overlapped MTAs, each for at least two of the three rusts. The number of MTAs for each MQTL also varied, ranging from 1 MTA (for several MQTLs) to 78 MTAs (for only one MQTL, namely MQTL6A.9) (Table S7).

MQTLs carrying R genes

R genes overlapping MQTLs were also identified; 44 R genes (10 Lr genes: 6 Sr genes and 28 Yr genes) were co-localized with 20 MQTLs; the number of R genes per MQTL could be as high as 13, sometimes including R...
genes for more than one rusts. For instance, MQTL2B.5 co-localized with the following 13 R genes including R genes for all the three rusts: Lr35, Sr36, YrSP, Yr5, Yr7, Yr43, Yr44, Yr53, Yr72, Lr13, Yr27, Sr9, and Sr28 genes; (Table S5).

**Candidate genes (CGs) and their gene ontology (GO) terms**

As many as 1,735 putative CGs were identified; these CGs belonged to 77 of the 78 MQTLs (MQTL4B.3 being the only...
exception). These CGs revealed several GO terms involved in widely known biological, molecular, and cellular processes. Some of the important GO terms included those involved in processes like the following: defense response, toxin activity, DNA binding, phosphorylation, protein ubiquitination, proteolysis, transmembrane transport, oxidation–reduction processes, catalytic activity, ATP binding, protein binding, heme binding, iron ion binding, metal ion binding, transmembrane transporter activity, oxidoreductase activity, etc. (Table S8).

**Differentially expressed CGs (DECGs) in MQTL regions**

In silico expression analysis was carried out for 541 CGs belonging to only 27 MQTLs, each with QTLs for all the three rusts. Only 81 CGs belonging to 22 MQTLs were differentially expressed. These 81 DECGs were also analyzed for GO enrichment (Fig. 6a). GO terms were not available for 14 of these genes. The DECGs encoded proteins belonging to the following categories: R-domain containing proteins, transcription factors (Zn finger binding proteins, SANT/Myb domains, NAC domain, bHLH TFs), transporters (ABC transporter, proton/oligo-peptide transporter, MFS transporter domain), protein kinases, proteins involved in calcium signaling, peptidases, proteins involved in oxidoreductive processes (RuBisCO, G6P_DH, FAD/NAD(P)-binding domain), etc. (Tables S9 and S10). Interestingly, 8 out of the 81 DECGs showed differential expression for all the three rusts. A representative heat map of these DECGs is presented in Fig. 6b.

**Homoeology among MQTLs**

Homoeologous relationships (including partial homoeology) among genes belonging to all the 78 MQTLs were also worked out. The data for number of total genes available in individual MQTL regions and those exhibiting homoeology are summarized in Fig. 7 and Table S11.
Table 2  MQTLs associated with multiple disease resistance identified in the present study

| Chr | MQTL/s (CI, in cM) | Flanking markers | QTLs involved (avg. LOD score; avg. PVE) |
|-----|---------------------|------------------|----------------------------------------|
| (a) Leaf rust, stem rust and yellow rust |
| 1A  | MQTL1A.1 (59.93–61.88) | IWB7590/AX-109017398 | 4 (4.67; 10.72) |
| 1B  | MQTL1B.1 (1.72–2.59) | AX-94847267/RAC875_c61512_173 | 6 (8.00;15.95) |
| 1D  | MQTL1D.2 (22.00–22.93); MQTL1D.6 (72.15–72.21) | AX-110480216/AX-11085909; IW82815/IW22866 | 3 (3.77; 22.72); 4 (5.12; 8.79) |
| 2A  | MQTL2A.3 (84.99–85.72); MQTL2A.4 (88.71–89.52); MQTL2A.5 (96.43–97.32) | wsnp_Ex_rep_c93362_82371891/Kukri_c84087_154;Excalibur_c92241_336/BobWhite_c2002_100;wsnp_Ex_c5412_9564478/GENE-1031_48 | 20 (11.36; 17.95); 3 (5.92; 14.69); 5 (11.93; 27.02) |
| 2B  | MQTL2B.1 (37.76–38.97); MQTL2B.2 (77.74–77.81); MQTL2B.3 (88.64–88.68) | AX-109493327/AX-109949983; AX-109001452/Kukri_s110874_162 | 14 (9.75; 24.30); 14 (15.50; 26.34); 35 (11.54; 21.39) |
| 2D  | MQTL2D.1 (9.13–11.03); MQTL2D.2 (32.54–34.78) | AX-94395108/Kukri_c5252_107 | 10 (5.91; 13.90); 13 (6.78; 20.02) |
| 3A  | MQTL3A.1 (21.06–21.76) | AX-94395108/Kukri_c5252_107 | 7 (5.08; 10.16) |
| 3B  | MQTL3B.1 (12.86–13.06) | AX-109995200/AX-109881148 | 39 (9.56; 18.71) |
| 3D  | MQTL3D.1 (5.21–7.93) | AX-94395108/Kukri_c5252_107 | 12 (5.27; 9.50); 3 (6.48; 14.87) |
| 4A  | MQTL4A.1 (18.02–19.62) | AX-109995200/AX-109881148 | 5 (14.74; 26.88); 7 (5.01; 11.53) |
| 5B  | MQTL5B.1 (64.30–66.33); MQTL5B.5 (157.42–162.48) | Xwmc734/AX-11001452/Kukri_s110874_162 | 12 (12.92; 28.12); 5 (5.35; 11.72) |
| 5D  | MQTL5D.1 (18.20–20.68); MQTL5D.2 (34.73–36.80) | AX-94942472/AX-94503507; AX-94487193/AX-95138970; GENE-3383_710/Ex_c29928_1020 | 12 (6.60; 9.54); 3 (5.70; 8.38); 4 (5.54; 17.10) |
| 6B  | MQTL6B.2 (287.60–287.64) | Xwmc388.3/AX-94463796 | 15 (4.61; 10.42) |
| 7A  | MQTL7A.1 (87.18–89.89) | AX-95190652/AX-109398226 | 4 (18.00; 37.32) |
| 7B  | MQTL7B.2 (53.57–54.31) | AX-95190652/AX-109398226 | 15 (5.15; 11.70) |
| 7D  | MQTL7D.1 (49.34–51.88); MQTL7D.2 (122.25–122.53) | AX-115515132/AX-11453717; IW82815/IW22866 | 25 (16.00; 29.56); 7 (14.79; 24.42) |
| (b) Leaf rust and yellow rust |
| 1B  | MQTL1B.1 (45.58–46.21); MQTL1B.4 (80.57–83.64); MQTL1B.5 (119.74–121.47); MQTL1B.6 (165.11–169.98) | IWB12256/AX-94409524; IWB12157/AX-95259256; IWB12619/AX-95259256; IWB12619/AX-19881148 | 3 (5.03; 14.77); 4 (5.41; 10.31); 13 (12.92; 28.12); 5 (5.35; 11.72) |
| 2D  | MQTL2D.3 (47.86–48.34) | AX-95205079/IWB42682 | 6 (3.45; 6.10) |
| 3A  | MQTL3A.2 (68.62–69.56) | AX-95205079/IWB42682 | 6 (3.45; 6.10) |
| 4B  | MQTL4B.5 (67.24–67.72) | AX-95205079/IWB42682 | 6 (3.45; 6.10) |
| 5B  | MQTL5B.3 (101.08–103.06); MQTL5B.4 (129.34–130.98); AX-94942472/AX-95403507; AX-947891/AX-10949983 | AX-95205079/IWB42682 | 12 (6.60; 9.54); 3 (5.70; 8.38); 4 (5.54; 17.10) |
| 6B  | MQTL6B.2 (287.60–287.64) | AX-95205079/IWB42682 | 12 (6.60; 9.54); 3 (5.70; 8.38); 4 (5.54; 17.10) |
| 7B  | MQTL7B.2 (53.57–54.31) | AX-95205079/IWB42682 | 12 (6.60; 9.54); 3 (5.70; 8.38); 4 (5.54; 17.10) |
| (c) Leaf rust and stem rust |
| 1D  | MQTL1D.1 (7.16–8.09); MQTL1D.3 (25.80–25.84); MQTL1D.5 (46.10–47.46) | Xmgw68/AX-110910133; AX-11147592/Xbarc152; XgII/IWA3124 | 2 (14.12; 23.13); 2 (5.40; 15.70); 4 (7.63; 20.04) |
| 3A  | MQTL3A.2 (68.62–69.56) | AX-109441469/AX-95135336 | 7 (5.71; 10.50) |
| 4B  | MQTL4B.5 (67.24–67.72) | AX-109441469/AX-95135336 | 7 (5.71; 10.50) |
| Chr | MQTL/s (CI, in cM) | Flanking markers | QTLs involved (avg. LOD score; avg. PVE) |
|-----|------------------|----------------|---------------------------------------|
| 5A  | MQTL5A.1 (69.56–71.38) | IWB20566/AX-94493739 | 6 (3.55; 6.32) |
| 5D  | MQTL5D.3 (93.03–99.12) | AX-108767468/AX-110967183 | 4 (9.77; 24.91) |
| 7A  | MQTL7A.2 (109.84–111.93) | AX-108833832/AX-108745312 | 5 (12.72; 15.22) |
| (d) Yellow rust and stem rust |
| 1A  | MQTL1A.2 (78.79–92.24); MQTL1A.4 (151.43–151.91) | wsnp_Ku_c5756_10191339/AX-109017398; Excalibur_rep_c110054_341/1132858 | 9 (6.05; 13.51); 3 (3.09; 8.84) |
| 1B  | MQTL1B.2 (31.19–32.10) | IWB11262/AX-94425009 | 7 (12.78; 19.99) |
| 2A  | MQTL2A.2 (58.97–59.63); MQTL2A.6 (114.15–114.78) | BS00068050_51/Kukri_c36139_292; AX-111707919/RAC875_c21013_1187 | 5 (5.77; 18.32); 11 (9.75; 15.54) |
| 2B  | MQTL2B.4 (103.74–104.30); MQTL2B.5 (156.51–156.65) | IWB9006/IWB12154; 2B_310443339/Xwmc317a | 16 (8.60; 18.69); 5 (9.44; 29.14) |
| 3B  | MQTL3B.2 (31.59–32.53); MQTL3B.3 (41.10–42.01) | AX-111494658/AX-111637887; AX-111684042/AX-111757878 | 7 (6.60; 14.21); 3 (5.17; 10.98) |
| 4A  | MQTL4A.3 (145.29–146.34) | IWB56172/Xbarc78 | 7 (18.62; 14.22) |
| 4B  | MQTL4B.4 (54.62–56.53); MQTL4B.8 (110.18–112.18) | AX-111719842/IWB6643; AX-109366086/RAC875_c10772_61 | 2 (9.76; 27.84); 5 (4.42; 5.74) |
| 4D  | MQTL4D.1 (0.03–0.05) | AX-111688098/AX-110768844 | 3 (5.85; 13.16) |
| 5B  | MQTL5B.2 (72.67–74.58) | AX-95205468/IWA8391 | 6 (4.68; 8.38) |
| 6A  | MQTL6A.3 (11.14–11.76); MQTL6A.4 (23.33–24.19); MQTL6A.5 (36.54–37.09); MQTL6A.6 (47.76–48.67); MQTL6A.8 (116.48–118.32) | IWB11274/AX-110585473; 6A_16883183/Xwmc201; Xgwm427/Xbarc3; 6A_83918914/IWB61322; Xeef80/IWB22389 | 4 (9.47; 31.99); 4 (28.05; 27.15); 5 (4.54; 6.78); 3 (6.03; 11.00); 3 (7.93; 14.82) |
| 7B  | MQTL7B.1 (44.85–44.22); MQTL7B.4 (138.35–152.65) | IWA1181/IWA7083; 1,112,830/Marker66313 | 18 (4.63; 10.84); 2 (5.60; 11.35) |
Discussion

In the past, disease resistance in crops was largely treated as a qualitative trait, where an individual R gene controls the resistance against a particular race of a pathogen following the widely known gene-for-gene relationship (Flor 1942, 1971). These R genes have also been pyramided involving several genes either for the same disease providing broad spectrum resistance for the same disease or for multiple diseases, providing MDR. More recently, with the availability of DNA-based molecular markers and statistical tools, QTL analysis (involving both interval mapping and GWAS) could be used for the identification of markers associated with QTLs for major individual diseases (Saini et al. 2022b). In some cases, markers have also been developed for MDR involving more than one
disease. A beginning has also been made in using associated markers for MAS for resistance against diseases in several crops including wheat (Kaur et al. 2020; Gupta et al. 2021; Sharma et al. 2021). Examples are also available for achieving MDR against more than one diseases, either through use of more than one gene/QTL or through the use of complex/pleiotropic loci (Table S12). We believe that if markers associated with QTLs or MQTLs, each carrying resistance for more than one disease are available, then it would be possible to transfer individual QTLs or MQTLs for MDR using only one set of associated markers for MAS.

The present study is an effort, where MQTLs were identified for MDR involving resistance to all the three rusts (LR, SR, and YR). In the published literature, the following are the only three studies, where MQTLs for MDR were identified: (i) in barley, for powdery mildew, net blotch, spot blotch, leaf blotch, brown rust, etc. (Schweizer and Stein 2011); (ii) in rice, for the blast, sheath blight and bacterial leaf blight (Kumar and Nadarajah 2020), and (iii) in maize, for northern leaf blight, gray leaf spot, and southern leaf blight (Ali et al. 2013). In wheat, the only other study for identification of MQTL for MDR was our own study involving the following five diseases: septoria tritici blotch, septoria nodorum blotch, fusarium head blight, Karnal bunt, and loose smut (Saini et al. 2022a). In the last study, among the five diseases used, fusarium head blight and septoria nodorum blotch are examples of diseases caused by necrotrophs, having very destructive pathogenesis strategies resulting in extensive necrosis, tissue maceration, etc., while Karnal bunt and septoria tritici blotch are caused by hemi-biotrophs, which follow a hemibiotrophic lifestyle, with a long symptomless, biotrophic phase, followed by a quick switch to necrotrophy associated with host necrosis and loose smut is caused by a biotroph which establish a long-term feeding relationship with the living cells of their hosts, rather than destroying/killing the host cells as part of the infection process. In contrast, the three rusts used in the present study are each caused by biotrophic pathogens. This suggested that MDR operates against all the different types of diseases in diverse host plants.

In the earliest review on MDR (for legumes), MDR was defined simply as “host-plant resistance to two or more diseases” (Nene 1988). In a relatively recent review, however, four different mechanisms for MDR were described for achieving MDR (Wiesner-Hanks and Nelson 2016; Fig. 8). Among these four methods, pyramiding or stacking of genes is the simplest method and has been successfully utilized in the past through conventional breeding. However, if we have markers associated with complex loci or QTLs
imparting resistance to multiple diseases, it will certainly help the breeders. Following are the four examples of such complex loci each involving two or more rusts in wheat: (i) *Lr34/Yr18/Sr57/Pm38Ltn1* (7DS; Krattinger et al. 2009), (ii) *Lr46/Yr29/Sr58/Pm39Ltn2* (1BL; Huerta-Espino et al. 2020), (iii) *Sr2/Lr27/Yr30/Pbc1* (3BS; Mago et al. 2011), (iv) *Lr67/Yr46/Sr55/Pm46/Ltn3* (4DL; Herrera-Foessel et al. 2014). These loci have often been described as pleiotropic, although evidence for pleiotropy against close linkage is not unequivocal.

Among the 1,146 QRLs (quantitative resistance loci; often described as QTLs) reported in 150 interval mapping studies that were used in the present study, most QRLs were each focused on single rust, but few studies also involved QRLs, each conferring resistance to two or all the three rusts suggesting the occurrence of MDR loci in some current wheat cultivars (Bemister et al. 2019; Prins et al. 2011; Singh et al. 2013; for details, see Table S2). Availability of individual MQTLs each for resistance to two or all the three rusts, as observed in the present study and reported in three
other crops (listed above), is yet another evidence in support of the MDR hypothesis (Wiesner-Hanks and Nelson 2016).

Of the 1,146 available QTLs, only 718 QTLs were projected onto the consensus map. The remaining 428 QTLs could not be projected owing to either of the following reasons: (i) they lacked common flanking markers between initial and consensus maps and (ii) they had comparatively large CI. The proportion of QTLs (62.65%) used for projection in the present study is higher than those in one of the two earlier studies on MQTL analysis for individual rusts (LR and YR) in wheat, where only 44.03% QTLs were projected (Soriano and Royo 2015). In the other study, 60.62% QTLs were projected (Jan et al. 2021), which is not very different from the present study. A higher proportion of projected QTLs in the present study may be attributed to the availability of more detailed information about many more QTLs and to the use of ultra-high density consensus map during the present study. The 86 MQTLs detected during the current study were obtained from 596 QTLs, leaving 122 unassigned QTLs or singletons. These 122 QTLs along with those which could not be projected on to the consensus map (despite having high LOD and PVE values) might be the unique loci, which may also be considered for breeding programmes for individual rusts. In the present study in wheat, a roughly seven-fold reduction in redundancy (86/596) for the genomic regions conferring resistance to LR, SR, and YR was observed. This observation is in sharp contrast to several earlier meta-QTL studies, where only 3 to five-fold reductions in redundancy of genomic regions were reported; these earlier studies include meta-QTL analysis for LR (Soriano and Royo, 2015), SR (Jan et al. 2021), fusarium head blight (Venske et al. 2019) and tan spot (Liu et al. 2020).

The frequency of MQTLs for all the three rusts among all the MQTLs in the present study (> 32%) is much higher than 5–10% MTAs for MDR reported in naturally occurring cultivars (Friesen et al. 2008; Gurung et al. 2009; Miedaner et al. 2020 Table S1). The MQTLs for MDR identified in the present study and the QTLs for MDR reported in naturally occurring cultivars can be used to provide resistance against any of the two or all the three rusts in wheat. There are at least two earlier studies, where QTLs for resistance against more than one disease have been identified in the same study. In one of these studies, 13 QTLs were reported to be significantly associated with resistance to four different diseases, namely, LR, YR, tan spot, and Karnal bunt (Singh et al. 2012). In the other study, a QTL representing a cluster of tightly linked loci on chromosome 3D for resistance to several foliar diseases (STB, tan spot, LR, SR, and YR) was reported (Zwart et al. 2010). Some of these naturally occurring QTLs for MDR, which correspond to MQTLs for MDR identified in the present study include the following: QSr.sun-3D, QStb.wai-3D, QRlnn.lrc-3D, QYls.lrc-3D, QTs.cimmyt-3AS, QYr.cimmyt-2AS (Zwart et al. 2010; Singh et al. 2012). It is thus apparent that only a small fraction of MDR MQTLs identified during the present study are currently known to occur in nature, thus underlining the importance of the present study.
MDR at the level of individual QTLs/genes or MQTLs (identified in the present study) may represent either several tightly linked loci or individual loci, each pleiotropic in nature. In case of closely linked multiple QTLs, these QTLs may be available either in the coupling phase or in the repulsion phase, resulting in positive and negative correlations between resistance to more than one disease. For instance, the wheat Sr2 locus, which confers resistance to LR, SR, and powdery mildew (Mago et al. 2011), was tightly linked in the repulsion phase to the Fhb1 locus, which confers resistance to fusarium head blight (Flemming 2012). In another study, QRLs for STB and yellow leaf spot inherited from one parent were linked in repulsion to the Lr24/Sr24 locus conferring resistance to leaf rust and stem rust inherited from the other parent (Zwart et al. 2010). The QTLs linked in the coupling phase can be readily introgressed together to provide resistance to two or more diseases, but introgression of the QTLs linked in the repulsion phase may be a trade-off, because the transfer of resistance to one disease is associated with susceptibility to another rust. As a result, it is obvious that in order to make effective use of a resistance source, a thorough understanding of the complexity of its inheritance is necessary.

The co-localization of ~69% of the physically anchored MQTLs with known GWAS-MTAs also deserves attention. Such a comparison of MQTLs with GWAS-MTAs in earlier studies reported the following proportion of MQTLs that could be verified by GWAS-MTAs: (i) 38.7% (Aduragbemi and Soriano 2021); (ii) 51.6% (Pal et al. 2021); (iii) 61.4% (Yang et al. 2021); (iv) 63.3% (Saini et al. 2022a), (v) 78.6% (Saini et al. 2021), (vi) 54.61% (Saini et al. 2022c), (vii) 46.3% (Gudi et al. 2021), (viii) 90.5% (Kumar et al. 2021). Our results fall within the range of per cent co-localization of MQTLs and the GWAS-MTAs in the above earlier studies. These co-localized MQTLs provide a basis for accurate mining of high confidence CGs associated with MDR in wheat.

Some MDR MQTLs detected during the current study also overlap the known rust resistance R genes (Table S5) which included 7 cloned genes (YrSP, Yr5, Yr7, Lr67/Yr46/ Sr35, Sr21, Sr33, and Yr36; Zhang et al. 2020). These genomic regions are believed to be involved in controlling both qualitative and quantitative resistance, making them important targets for introgression into susceptible wheat lines in order to improve resistance to the three rusts. Following are some examples:

(i) MQTL2A.6 overlaps the following two resistance genes: Sr21, that encodes a coiled-coil nucleotide-binding leucine-rich repeat (NLR) protein and confers resistance against races belonging to Ug99 group at high temperature (Chen et al. 2018), and Yr32, which confers resistance against YR, effective both at the seedling and adult growth stage (Eriksen et al. 2004).

(ii) MQTL2B.5 is associated with two Lr genes (Lr13 and Lr35), three Sr genes (Sr9, Sr28 and Sr36), and eight Yr genes (Yr5, Yr7, Yr27, Yr43, Yr44, Yr53, Yr72 and YrSP), making this region a hotspot that can be used for introgression of MDR in wheat. Among three Sr genes associated with MQTL2B.5, intriguingly Sr9 gene seems to be interesting, because it is known to have the following eight different alleles: Sr9a, Sr9b, Sr9c, Sr9d, Sr9e, Sr9f, Sr9g, and Sr9h; this demonstrates occurrence of alleles with unique race specificities to SR races; among these, Sr9c and Sr9h are effective against most destructive SR race, Ug99 race (Rouse et al. 2014). Among the 13 R genes that are co-localized with MQTL2B.5, three R genes, namely, Yr5, Yr7, and YrSP have recently been cloned (Marchal et al. 2018); Yr5, which remains effective to a broad range of Pst isolates worldwide, is closely related yet distinct from Yr7, whereas YrSP is a truncated version of Yr5 with 99.8% sequence identity (Marchal et al. 2018). These three Yr genes are members of a complex resistance gene cluster on chromosome 2B that encodes an NLR protein with a non-canonical N-terminal zinc-finger BED domain that differs from that found in non-NLR wheat proteins (Marchal et al. 2018). Diagnostic markers have also been developed for the above three Yr genes; similar markers can also be developed for other non-allelic Lr and Sr genes which may accelerate the haplotype analysis and expedite stacking of different genes through MAS.

(iii) MQTL2D.1 is co-localized with a seedling LR resistance gene (Lr15) (Dholakia et al. 2013), two adult plant YR resistance genes (Yr16, Yr54) and an all-stage YR resistance gene (Yr55) (Rani et al. 2019).

(iv) MQTL3A.1 overlap with Lr63, Yr76; among these, Lr63 is responsible for low to intermediate infection types to most P. triticina isolates, however in combinations with other effective seedling or AP resistance genes, it can be used to develop wheat cultivars with highly effective LR resistance.

(v) MQTL4D.3 is associated with Lr67 (part of a complex locus Lr67/Yr46/Sr55/Pm46/Ltn3 that has been cloned); the gene is associated with co-segregation of five diseases (LR, YR, SR, Pm and leaf tip necrosis (Herrera-Foessel et al. 2014; Moore et al. 2015). The gene encodes a hexose transporter (LR67res), which differs from the susceptible form of the same protein (LR67sus) by two amino acids conserved across the orthologous hexose transporters. The protein LR67res and related proteins encoded by homeoalleles function as high-affinity glucose transporters. Through heterodimerization with these functional transporters, LR67res shows a dominant-negative effect on glucose uptake. Changes in hexose transport in infected leaves could
explain the plant’s ability to suppress the growth of multiple biotrophs (Moore et al. 2015).

The MDR MQTLs, which co-localize with known Lr, Sr, and Yr genes, may also be important targets for introgression into susceptible wheat lines for enhancing the resistance against all the three rusts. Although there are hardly any examples of transfer of MDR QTL for all three rusts, examples for transfer of multiple QRLs for the same disease are available. For instance, Hu et al. (2020) transferred two QRLs (Qyr.nafu-2BL and QYr.nafu-3BS) for resistance to SR from wheat line P9897 into three Chinese elite wheat cultivars, Chuanmai 42, Xiangmai 25, and Zhengmai 9023. They concluded that a combination of major known gene and QRLs may widen the resistance spectrum and enhance the resistance.

There are also examples, where CGs underlying MQTLs were identified for several traits including drought tolerance (Kumar et al. 2020a), tan spot resistance (Liu et al. 2020), and fusarium head blight resistance (Venske et al. 2019) in wheat. For identification of CGs, we used a strategy earlier used by Saini et al. (2022c) in which the physical positions of MQTL peaks were determined, and then, the 1 Mb intervals on either side of the MQTL peaks were considered for the identification of CGs available in MQTL regions. The results of the present study involving 1,735 CGs could also be examined in the context of MDR; at least 285 of these CGs are known to encode different R domains including NBS-LRR, coiled coil, NB-ARC, protein kinases domains (e.g., serine/threonine-protein kinase), WRKY motif, etc. The number of CGs encoding any of the R domains ranged from one CG available from each of several MQTLs (e.g., MQTL1B.3, 1B.5, 2A.4, 2D.1, etc.) to a maximum of 28 CGs available from MQTL5D.3 with an average of around 5 CGs per MQTL.

The 81 DECGs identified during the present study (Table S9 and S10) encode proteins carrying the following domains: kinase domain, NBS-LRR domain, serine/threonine-protein kinase, UDP-glycosyltransferase, WRKY domains, F-box domain, glycosyl hydrolase, ABC transporter-like domain, WD40-repeat-containing domain, and MFS transporter. Following are some examples of known genes representing CGs, with a role in disease resistance: (i) Serine/threonine protein kinase (STPK-V) gene, which is a member of Pm21 family for resistance against powdery mildew (Cao et al. 2011; Xing et al. 2018). (ii) genes encoding proteins with NBS-LRR domains; these genes resemble cloned Yr genes like Yr10, Yr5, etc. (Liu et al. 2014; Marchal et al. 2018); (iii) genes like Sr21 for resistance to stem rust in wheat (Chen et al. 2018) and Rpg5 in barley (Brueggeman et al. 2008), which encode proteins with nucleotide-binding-site, leucine-rich, and protein kinase domains. (iv) Yr genes like Yr46 which was shown to encode for hexose transporter (Moore et al. 2015). (v) genes encoding UDP-glucosyltransferases that were earlier reported to show differential expression due to SR infection in wheat genotypes indicating their role in Yr39 mediated SR resistance (Coram et al. 2008). (vi) WRKY domain containing genes that were recently found to encode proteins like those encoded by cloned YrU gene (Wang et al. 2020). (vii) F-box domain containing gene that was identified as a CGs underlying the YrR39 locus in wheat and was shown to be upregulated due to SR infection (Yin et al. 2018).

**Conclusions**

In the present study, we integrated the results of QTL mapping studies on LR, SR, and YR resistance leading to the identification of 86 MQTLs. More than half of these MQTLs were validated using GWAS results. Some of these MQTLs were found to be co-located with as many as 44 known major resistance R genes. Further, 28 MQTLs provided resistance to all the three rusts, while each of other 43 MQTLs provided resistance to any of the two rusts. Putative CGs were identified and 81 CGs showed differential expression, encoding important proteins. Eight DECGs showed differential expression for all the three rusts. Ten promising MDR-MQTLs were recommended for use in marker-assisted breeding for the development of resistance to three rust diseases in wheat cultivars. This study can also help better define the various mechanisms associated with MDR in wheat.

**Supplementary Information** The online version contains supplementary material available at https://doi.org/10.1007/s00122-022-04119-7.

**Acknowledgements** Thanks are due to Department of Biotechnology (DBT), Government of India, for financial support (BT/PR21024/AGIII/103/925/2016) and (BT/NABI-Flagship/2018). Thanks are also due to Indian National Science Academy (INSA), New Delhi, for the award of the positions of INSA-Senior Scientist and INSA Honorary Scientist to HSB.

**Author Contribution statement** PKG, HSB, PKS, and SK conceived and planned this study, NP, IJ, AK, and KK collected the literature and tabulated the QTL data for meta-QTL analysis. NP, IJ, DKS prepared the input files and performed QTL projection and meta-QTL analysis. NP, IJ and DKS interpreted the results and wrote the first draft of the manuscript. PKG, HSB, PKS, and SK edited and finalized the manuscript with the help of NP, IJ and DKS.

**Funding** No funding was received from any source for conducting the present study.

**Availability of data and material** Additional data relevant to this paper are available in the form of supplementary material.
Declarations

Conflict of interest  The authors declare no conflicts of interest.

References

Aduragbemi A, Soriano JM (2021) Unravelling consensus genomic regions conferring leaf rust resistance in wheat via meta-QTL analysis. BMC Genom. https://doi.org/10.1186/s12862-021-01557-0

Afzal SN, Haque ML, Ahmedani MS, Bashir S, Rattu AR (2007) Assessment of yield losses caused by Puccinia striiformis triggering stripe rust in the most common wheat varieties. Pak J Bot 39:2127–2134

Agenbag GM, Pretorius ZA, Bender CM, MacCormack R, Prins R (2014) High-resolution mapping and new marker development for adult plant stripe rust resistance QTL in the wheat cultivar Kariega. Mol Breed 34:2005–2020

Alemu A, Feyissa T, Maccaferri M, Sciara G, Tuberosa R, Ammar K, Badebo A, Avedo M, Letta T, Ayebay B (2021) Genome-wide association analysis unveils novel QTLs for seminal root system architecture traits in Ethiopian durum wheat. BMC Genomics 22:1–16

Ali F, Pan Q, Chen G, Zahid KR, Yan J (2013) Evidence of multiple disease resistance (MDR) and implication of meta-analysis in marker assisted selection. PLoS ONE 8:e68150

Arcade A, Labourette A, Falque M, Mangin B, Chardon F, Charcosset A, Joets J (2006) BioMercator: integrating genetic maps and QTL towards discovery of candidate genes. Bioinformatics 22:2324–2326

Ausems ER (1943) Breeding for disease resistance in wheat, oats, barley and flax. Bot Rev 9:207–260

Bajgain P, Rouse MN, Tsilo TJ, Macharia GK, Bhavani S, Jin Y, Anderson JA (2016) Nested association mapping of stem rust resistance in wheat using genotyping by sequencing. PLoS ONE 11:e0155760

Bemister DH, Semagn K, Iqbal M, Randhawa H, Strelkov SE, Spaner DM (2019) Mapping QTL associated with stripe rust, leaf rust, and leaf spotting in a Canadian spring wheat population. Crop Sci 59:650–658

Brueggeman R, Druka A, Nirmala J, Cavileer T, Drader T, Rostoks N, Mirlohi A, Bennypaul H, Gill U, Kudrna D, Whitelaw C (2008) The stem rust resistance gene Rpp5 encodes a protein with nucleotide-binding-site, leucine-rich, and protein kinase domains. Proc Natl Acad Sci 105:14970–14975

Cao A, Xing L, Wang X, Yang X, Wang W, Sun Y, Qian C, Ni J, Chen Y, Liu D, Wang X (2011) Serine/threonine kinase gene Stpk-V, a key member of powdery mildew resistance gene Pm211, confers powdery mildew resistance in wheat. Proc Natl Acad Sci 108:7727–7732

Cavanagh CR, Chao S, Wang S, Huang BE, Stephen S, Kiani S, Forrest K, Sainenc T, Brown-Guedira GL, Akhunova A, See D (2013) Genome-wide comparative diversity uncovers multiple targets of selection for improvement in hexaploid wheat landraces and cultivars. Proc Natl Acad Sci 110:8057–8062

Chardon F, Vilron B, Moreau L, Falque M, Joets J, Decousset L, Murigneux A, Charcosset A (2004) Genetic architecture of flowering time in maize as inferred from quantitative trait loci meta-analysis and synteny conservation with the rice genome. Genetics 168:2169–2185

Chen XM (2005) Epidemiology and control of stripe rust [Puccinia striiformis f. sp. triticici] on wheat. Can J Plant Pathol 27:314–337

Chen S, Zhang W, Bolus S, Rouse MN, Dubcovsky J (2018) Identification and characterization of wheat stem rust resistance gene Sr2l effective against the Ug99 race group at high temperature. PLoS Genet 14:e1007287

Cloutier S, Wang Z, Banks TW, Jordan MC, McCallum BD (2008) Gene expression of plant defence pathways using Lrl transgenic lines and the Affymetrix wheat Chip. In: The 11th international wheat genetics symposium proceedings edited by Rudi Appels Russell Eastwood Evans Lagudah Peter Langridge Michael Mackay Lynne. Sydney University Press

Coram TE, Settles ML, Chen X (2008) Transcriptome analysis of high-temperature adult-plant resistance conditioned by Yr39 during the wheat–Puccinia striiformis f. sp. tritici interaction. Mol Plant Pathol 9:479–493

Cui F, Zhang N, Fan XL, Zhang W, Zhao CH, Yang LJ, Pan RQ, Chen M, Han J, Zhao XQ, Ji J (2017) Utilization of a Wheat 660K SNP array-derived high-density genetic map for high-resolution mapping of a major QTL for kernel number. Sci Rep 7:1–12

Darvasi A, Soller M (1997) A simple method to calculate resolving power and confidence interval of QTL map location. Behav Genet 27:125–132

Dholakia BB, Rajwade AV, Hosmani P, Khan RR, Chavan S, Reddy DMR, Lagu MD, Bansal UK, Saini RG, Gupta VS (2013) Molecular mapping of leaf rust resistance gene Lr15 in hexaploid wheat. Mol Breed 31:743–747

Dobson A, Bunting DC, Cabrera-Quio LE, Uaay C, Saunders DG (2016) The host-pathogen interaction between wheat and yellow rust induces temporally coordinated waves of gene expression. BMC Genom 17:1–14

Edae EA, Rouse MN (2020) Association mapping of resistance to emerging stem rust pathogen races in spring wheat using genotyping-by-sequencing. The Plant Genome 13:e20050

Endelman JB, Plomion C (2014) L.Pmerge: an R package for merging genetic maps by linear programming. Bioinformatics 30:1623–1624

Eriksen L, Afshari F, Christiansen MJ, McIntosh RA, Jahoor A, Wellings CR (2004) Yr32 for resistance to stripe (yellow) rust present in the wheat cultivar Carstens V. Theor Appl Genet 108:567–575

Fatima F, McCallum BD, Pozniak CJ, Hiebert CW, McCartney CA, Fedak G, You FM, Cloutier S (2020) Identification of new leaf rust resistance loci in wheat and wild relatives by array-based SNP genotyping and association genetics. Front Plant Sci 11:1728

Flemmig EL (2012) Molecular markers to deploy and characterize stem rust resistance in wheat. Dissertation, North Carolina State University

Flor HH (1942) Inheritance of pathogenicity in Melampsora lini. Physopathology 32:653–669

Flor HH (1971) Current status of the gene-for-gene concept. Annu Rev Phytopathol 9:275–296

Friesen TL, Xu SS, Harris MO (2008) Stem rust, tan spot, Stagonosporonudor blotch, and Hessian fly resistance in Langdon durum-Aegilops tauschi synsthesi hexaploid wheat lines. Crop Sci 48:1062–1070

Gao L, Turner MK, Chao S, Kolmer J, Anderson JA (2016) Genome-wide association study of seedling and adult plant leaf rust resistance in elite spring wheat breeding lines. PLoS One 11:e0148671

Genievskaia Y, Turuspekov Y, Rsaliev A, Abagalievia S (2020) Genome-wide association mapping for resistance to leaf, stem, and yellow rusts of common wheat under field conditions of South Kazakhstan. Peer J 8:e9820

Goffinet B, Gerber S (2000) Quantitative trait loci: a meta-analysis. Genetics 155:463–473

Gudi S, Saini DK, Singh G, Halladakeri P, Shamshad M, Tanin MJ, Kumar P, Sharma A (2021) Unravelling consensus genomic regions associated with quality traits in wheat (Triticum aestivum)
Kaur B, Bhathia D, Mavi GS (2021) Eighty years of gene-for-gene relationship and its applications in identification and utilization of R genes. J Genet 100:1–17

Knott DR (1989) The effect of transfers of alien genes for leaf rust resistance on the agronomic and quality characteristics of wheat. Euphytica 44:65–72

Kolmer JA, Garvin DF, Jin Y (2011) Expression of a Thatcher wheat adult plant stem rust resistance QTL on chromosome arm 2BL is enhanced by Lr34. Crop Sci 51:526–533

Kou Y, Wang S (2010) Broad-spectrum and durability: understanding of quantitative disease resistance. Curr Opin Plant Biol 13:181–185

Krattinger SG, Lagudah ES, Spielmeyer W, Singh RP, Huerta-Espino J, McFadden H, Bossolin E, Seltler LL, Keller B (2009) A putative ABC transporter confers durable resistance to multiple fungal pathogens in wheat. Science 323:1360–1363

Kumar IS, Nadarajah K (2020) A meta-analysis of quantitative trait loci associated with multiple disease resistance in rice (Oryza sativa L.). Plants 9:1491

Kumar A, Saripalli G, Jan I, Kumar K, Sharma PK, Balyan HS, Gupta PK (2020a) Meta-QTL analysis and identification of candidate genes for drought tolerance in bread wheat (Triticum aestivum L.). Physiol Mol Biol Plants 26:1713–1725

Kumar D, Kumar A, Chhokar V, Gangwar OP, Bhardwaj SC, Sivasamy M, Prasad SV, Prakash TL, Khan H, Singh R, Sharma P (2020b) Genome-wide association studies in diverse spring wheat panel for stripe, stem, and leaf rust resistance. Front Plant Sci 11:748

Kumar A, Saripalli G, Jan I, Kumar K, Sharma PK, Balyan HS, Gupta PK (2020c) Meta-QTL analysis and identification of candidate genes for drought tolerance in bread wheat (Triticum aestivum L.). Physiol Mol Biol Plants 26:1713–1725

Kumar S, Singh VP, Saini DK, Sharma H, Saripalli G, Kumar S et al (2021) Meta-QTLs, ortho-MQTLs, and candidate genes for thermotolerance in wheat (Triticum aestivum L.). Mol Breed 41:1–22

Lagudah ES, Krattinger SG, Herrera-Foessel S, Singh RP, Huerta-Espino J, Spielmeyer W, Brown-Guedira G, Seltler LL, Keller B (2009) Gene-specific markers for the wheat gene Lr34/Yr18/Pm38 which confers resistance to multiple fungal pathogens. Theor Appl Genet 119:889–898

Ledesma-Ramírez L, Solís-Moya E, Iturriaga G, Sehgal D, Reyes-Valdes MH, Montero-Taveras V, Sansaloni CP, Burgueno J, Ortiz C, Aguirre-Manchilla CL, Ramírez-Pimentel JG (2019) GWAS to identify genetic loci for resistance to yellow rust in wheat pre-breeding lines derived from diverse exotic crosses. Front Plant Sci 10:1390

Leonova IN, Skolotneva ES, Salina EA (2020) Genome-wide association study of leaf rust resistance in Russian spring wheat varieties. BMC Plant Biol 20:1–13

Letta T, Maccaferrì M, Badebo A, Ammar K, Ricci A, Crossa J, Tuberosa R (2013) Searching for novel sources of field resistance to Ug99 and Ethiopian stem rust races in durum wheat via association mapping. Theor Appl Genet 126:1237–1256

Li W, Deng Y, Ning Y, He Z, Wang GL (2020) Exploiting broad-spectrum disease resistance in crops: from molecular dissection to breeding. Annu Rev Plant Biol 71:575–603

Liu W, Frick M, Huel R, Nykiiforuk CL, Wang X, Gaudet DA, Eudes F, Conner RL, Kuzyk A, Chen Q, Kang Z (2014) The stripe rust resistance gene Yr10 encodes an evolutionary-conserved and unique CC–NBS–LRR sequence in wheat. Mol Plant 7:1740–1755

Liu Y, Salsman E, Wang R, Galagedara N, Zhang Q, Fiedler JD, Liu Z, Xu S, Faris J, Li X (2020) Meta-QTL analysis of tan spot resistance in wheat. Theor Appl Genet 133:2363–2375

Maccaferrì M, Zhang J, Bulli P, Abate Z, Chao S, Cantu D, Bossolin E, Chen X, Pumphrey M, Dubcovsky J (2015) A genome-wide association study of resistance to stripe rust (Puccinia striiformis)
...in a worldwide collection of hexaploid spring wheat (*Triticum aestivum* L.), *G3* Genes Genomes Genet 5(3):449–465.

Mago R, Tabe L, McIntosh RA, Pretorius Z, Kota R, Pauw E, Wicker T, Breen J, Lagudah ES, Ellis JG, Spielmeyer W (2011) A multiple resistance locus on chromosome arm 3BS in wheat confers resistance to stem rust (*Sr2*) leaf rust (*Sr27*) and powdery mildew. Theor Appl Genet 123:615–623.

Marchal C, Zhang J, Zhang P, Fenwick P, Steuernagel B, Adamski NM, Boyd L, McIntosh R, Wulf BB, Berry S, Lagudah E (2018) BED-domain-containing immune receptors confer diverse resistance spectra to yellow rust. Nat Plant 4:662–668.

Marone D, Russo MA, Laidi G, De Vita P, Papa R, Blanco A, Gadaleta A, Rubiales D, Mastrangelo AM (2013) Genetic basis of quantitative and qualitative resistance to powdery mildew in wheat: from consensus regions to candidate genes. BMC Genom 14:1–17.

McIntosh RA, Dubcovsky J, Rogers W, Morris C, Appels R, Xia X (2016) Catalogue of gene symbols for wheat: 2015–2016 supplement. Annual Wheat Newsletter 58:1–18.

Megerssa SH, Ammar K, Acevedo M, Brown-Guedira G, Ward B, Degete AG, Randhawa MS, Sorrells ME (2020) Multiple-race stem rust resistance loci identified in durum wheat using genome-wide association mapping. Front Plant Sci 11:1934.

Miedaner T, Rapp M, Flath K, Longin CFH, Würschum T (2020) Genetic architecture of yellow and stem rust resistance in a durum wheat diversity panel. Euphytica 215:1–17.

Miedaner T, Akel W, Flath K, Jacobi A, Taylor M, Longin F, Würschum T (2020) Molecular tracking of multiple disease resistance in a winter wheat diversity panel. Theor Appl Genet 133:419–431.

Moore JW, Herrera-Foessel S, Lan C, Schnippenkoetter W, Ayliffe JC, Doss R, Prins R, Pretorius ZA, Bender CM, Lehmensiek A (2011) QTL mapping of Stripe, leaf and stem rust resistance genes in a *Kariega x Avocet S* doubled haploid wheat population. Mol Breed 27:2767–2786.

Pal N, Saini DK, Chahal A, Srivastava P, Gupta PK (2022c) Meta-QTLs, ortho-QTLs and candidate genes for grain yield and associated traits in wheat (*Triticum aestivum* L.). Theor Appl Genet 135(3):1049–1081.

Pal N, Saini DK, Chahal A, Srivastava P, Gupta PK (2022a) Meta-analysis reveals consensus genomic regions associated with multiple disease resistance in wheat (*Triticum aestivum* L.). Mol Breed 42:1–2.

Pal N, Saini DK, Srivastava P, Pal N, Gupta PK (2022c) Meta-QTLs, ortho-QTLs and candidate genes for grain yield and associated traits in wheat (*Triticum aestivum* L.). Theor Appl Genet 135(3):1049–1081.

Paterson J (2014) Multiple disease resistance the holy grail. https://grdc.com.au/resources-and-publications/groundcover/ground-cover-supplements/gcs110/multiple-disease-resistance-the-holy-grail. Accessed 25 Jan 2019.

Pooja S, Sharma RB, Singh AK, Rakesh S, Sundee K (2014) Multiple disease resistance in wheat: need of today. Wheat Interv Serv 118:7–16.

Pradhan AK, Kumar S, Singh AK, Budhlakoti N, Mishra DC, Chauhan D, Mittal S, Grover M, Kumar S, Gangwar GP, Kumar S (2020) Identification of QTLs/defense genes effective at seedling stage against prevailing races of wheat stripe rust in India. Front Genet 11:572975.

Prins R, Pretorius ZA, Bender CM, Lehmensiek A (2011) QTL mapping of Stripe, leaf and stem rust resistance genes in a *Kariega x Avocet S* doubled haploid wheat population. Mol Breed 27:259–270.

Ramirez-Gonzalez RH, Borrill P, Lang D, Harrington SA, Brinton J, Venturini L, Davey M, Jacobs J, Van Ex F, Pasha A, Khediker Y (2018) The transcriptional landscape of polyploid wheat. Science 361:6403.
(2013) Identification and mapping of leaf, stem and Stripe rust resistance quantitative trait loci and their interactions in durum wheat. Mol Breed 31:405–418
Singh K, Batra R, Sharma S, Saripalli G, Gautam T, Singh R, Pal S, Malik P, Kumar M, Jan I, Singh S (2021) WheatQTLdb: a QTL database for wheat. Mol Genet Genom 296:1051–1056
Soko T, Bender CM, Prins R, Pretorius ZA (2018) Yield loss associated with different levels of stem rust resistance in bread wheat. Plant Dis 102:2531–2538
Somers DJ, Isaac P, Edwards K (2004) A high-density microsatellite consensus map for bread wheat (Triticum aestivum L.). Theor Appl Genet 109:1105–1114
Soriano JM, Royo C (2015) Dissecting the genetic architecture of leaf rust resistance in wheat by QTL MQTL analysis. Phytopathology 105:1585–1593. https://doi.org/10.1094/PHYTO-05-15-0130-R
Sosnowski O, Charcosset A, Joets J (2012) BioMercator V3: an upgrade of genetic map compilation and quantitative trait loci meta-analysis algorithms. Bioinformatics 28:2082–2083
Venske E, dos Santos RS, da Rosa FD, Rother V, Maia LC, Pegoraro C, Costa De Oliveira A (2019) MQTL analysis of the QTLome of Fusarium head blight resistance in bread wheat: refining the current puzzle. Front Plant Sci 10:727
Veyrieras JB, Goffinet B, Charcosset A (2007) Meta QTL: a package of new computational methods for the MQTL analysis of QTL mapping experiments. BMC Bioinformatics 8:49. https://doi.org/10.1186/1471-2105-8-49
Visscher PM, Goddard ME (2004) Prediction of the confidence interval of quantitative trait loci location. Behav Genet 34:477–482
Wang S, Wong D, Forrest K, Allen A, Chao S, Huang BE, Maccferri M, Salvi S, Milner SG, Cattivelli L, Mastrangelo AM (2014) Characterization of polyploid wheat genomic diversity using a high density 90000 single nucleotide polymorphism array. Plant Biotechnol J 12:787–796
Wang J, Li Z, Shi L, Zhu L, Ren Z, Li LD, Shah SJA (2015) QTL mapping for adult-plant leaf rust resistance genes in Chinese wheat cultivar Weimai 8. Czech J Genet Plant Breed 51:79–85
Wang H, Zou S, Li Y, Lin F, Tang D (2020) An ankyrin-repeat and WRKY-domain-containing immune receptor confers stripe rust resistance in wheat. Nat Commun 11:1–11
Wiesner-Hanks T, Nelson R (2016) Multiple disease resistance in plants. Annu Rev Phytopathol 54:229–252
Winfield MO, Allen AM, Burridge AJ, Barker GL, Benbow HR, Wilkinson PA, Coghill J, Waterfall C, Davassi A, Scopes G, Pirani A (2016) High-density SNP genotyping array for hexaploid wheat and its secondary and tertiary gene pool. Plant Biotechnol J 14:1195–1206
Xing L, Hu P, Liu J, Witek K, Zhou S, Xu J, Zhou W, Gao L, Huang Z, Zhang R, Wang X (2018) Pm21 from Haynaldia villosa encodes a CC-NBS-LRR protein conferring powdery mildew resistance in wheat. Mol Plant 11:874–878
Yadav IS, Sharma A, Kaur S, Nahar N, Bhardwaj SC, Sharma TR, Chhuneja P (2016) Comparative temporal transcriptome profiling of wheat near isogenic line carrying Lr57 under compatible and incompatible interactions. Front Plant Sci 7:1943
Yang Y, Amo A, Wei D, Chai Y, Zheng J, Qiao P, Cui C, Lu S, Chen L, Hu YG (2021) Large-scale integration of meta-QTL and genome-wide association study discovers the genomic regions and candidate genes for yield and yield-related traits in bread wheat. Theor Appl Genet 134:1–27
Yin JL, Fang ZW, Sun C, Zhang P, Zhang X, Lu C, Wang SP, Ma DF, Zhu YX (2018) Rapid identification of a stripe rust resistant gene in a space-induced wheat mutant using specific locus amplified fragment (SLAF) sequencing. Sci Rep 8:1–9
Yu LX, Morgounov A, Wanyera R, Keser M, Singh SK, Sorrells M (2012) Identification of Ugg99 stem rust resistance loci in winter wheat germplasm using genome-wide association analysis. Theor Appl Genet 125:749–758
Zegeye H, Rasheed A, Makdis F, Badebo A, Ogbonnaya FC (2014) Genome-wide association mapping for seedling and adult plant resistance to stripe rust insynthetic hexaploid wheat. PloS one 9:e105593
Zhang H, Yang Y, Wang C, Liu M, Li H, Fu Y, Wang Y, Nie Y, Liu X, Ji W (2014) Large-scale transcriptome comparison reveals distinct gene activations in wheat responding to stripe rust and powdery mildew. BMC Genom 15:1–14
Zhang P, Qi A, Zhou Y, Xia X, He Z, Li Z, Liu D (2016) Quantitative trait loci mapping of adult-plant resistance to leaf rust in a Fun-dula 900x‘Thatcher’wheat cross. Plant Breed 136:1–7
Zhang J, Zhang P, Dodds P, Lagudah E (2020) How target-sequence enrichment and sequencing (TeSeq) pipelines have catalyzed resistance gene cloning in the wheat-rust pathosystem. Front Plant Sci 11:678
Zhang P, Yan X, Gebrevahid TW, Zhou Y, Yang E, Xia X, He Z, Li Z, Liu D (2021) Genome-wide association mapping of leaf rust and stripe rust resistance in wheat accessions using the 90K SNP array. Theor Appl Genet 134:1233–1251
Zwart RS, Bansal UK, Thompson JP, Williamson PM, Bariiana HS (2008) QTL mapping of multiple disease resistance traits in a synthetic hexaploid x bread wheat population. In: Proceedings 11th international wheat genetics symposium, Brisbane, pp 1–3
Zwart RS, Thompson JP, Miligate AW, Bansal UK, Williamson PM, Raman H, Bariiana HS (2010) QTL mapping of multiple foliar disease and root-lesion nematode resistances in wheat. Mol Breed 26:107–124

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.