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Genetic dissection and genomic prediction of drought indices in bread wheat (*Triticum aestivum* L.) genotypes

Zakaria El Gataa [a,*], Alemu Admas [b], Samira El Hanafi [c], Zakaria Kehel [a], Fatima Ezzahra Rachdad [a], Wuletaw Tadesse [a]

[a] The International Center for Agricultural Research in the Dry Areas (ICARDA), Rabat, Morocco
[b] Department of Plant Breeding, Swedish University of Agricultural Sciences, 23422, Lomma, Sweden
[c] Leibniz Institute of Plant Genetics and Crop Plant Research (IPK), Gatersleben, Germany

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ABSTRACT

Drought constitutes the main obstacle to agricultural productivity in the Central and West Asia and North Africa (CWANA) region, notably leading to substantial reduction in wheat yields due to terminal water stress. The adoption of drought-resistant wheat varieties appears to be a vital strategy to maintain wheat production in the face of climatic challenges. In this context, a study was conducted utilizing a set of 198 elite bread wheat genotypes developed at the International Center for Agricultural Research in the Dry Areas (ICARDA). This set of elite genotypes was evaluated at the Sidi Al-Aidi station in Morocco over two years (2021–2022), under rain-fed and irrigated conditions. Phenotypic assessments for grain yield and drought indices were performed, alongside genotyping the population using 15k SNP markers. These preparatory steps facilitated a genome-wide association study (GWAS) and genomic prediction, leveraging the Mixed Linear Model (MLM) to pinpoint marker-trait associations (MTAs) and candidate genes pertinent to grain yield and drought indices. The results manifested substantial variations in both grain yield and drought indices among the genotypes tested. Grain yield performance ranged from 0.34 to 2.57 t/ha under rain-fed conditions and 1.12 to 4.57 t/ha under irrigated scenarios. The comprehensive analysis identified 39 significant MTAs ($p < 0.001$) and 14 putative genes associated with drought indices and grain yield. Noteworthy is the marker "*wsnp_Ex_c12127_19394952*" on chromosome 5B, which displayed a significant correlation with grain yield in rain-fed environments. Furthermore, the most prominent marker linked to tolerance index (TOL) was "BobWhite_*c42349_99*", situated on chromosome 5A and associated with the *TraesCS5A02G498000* gene. This gene plays a critical role, encoding for catalase protein crucial for response to hydrogen peroxide. These markers could be used for marker-assisted selection in wheat breeding programs targeting drought tolerance.

1. Introduction

Bread wheat holds a pivotal role as a staple food in both the Central and West Asia and North Africa (CWANA) and Sub-Saharan Africa (SSA) regions. However, the combination of limited water availability due to drought and the intensifying terminal heat stress take a toll on wheat, hindering their growth, development, and, ultimately, their ability to produce an optimal yield [1]. Over recent decades, the CWANA and SSA regions have experienced recurrent episodes of drought and heatwaves, which have constrained wheat production and underscored the urgent need for adaptive strategies [2]. Morocco harvested 5.06 million tons of bread wheat in season 2021–2022 on 3.2 million hectares, averaging 1.5 tons per hectare [3].

Drought tolerance indices are crucial tools in wheat breeding to identify and develop cultivars resistant to drought stress. These mathematically developed indicators consider several factors, including yield stability, biomass retention, and physiological traits such as stomatal conductance and root depth. Researchers and breeders may develop wheat varieties that can withstand arid and semi-arid environments by identifying genotypes that perform better under drought conditions by analyzing these parameters [4]. The use of drought tolerance indices also enables a more nuanced comprehension of how plants react to drought at various developmental stages, shedding light on the intricate interaction between genetic and environmental variables that determine drought

* Corresponding author.
E-mail address: elgataa.zakaria@gmail.com (Z. El Gataa).

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tolerance. As a result, these indices continue to serve as the cornerstone of efforts to increase the drought resistance of wheat crops, opening the path for sustainable agriculture in the face of climate change [5].

Drought stress orchestrates a complex interplay of mechanisms that are influenced by numerous factors, including the species of crops, the severity and duration of the drought, and the specific growth stages of the plant in question. Understanding these dynamics is pivotal in the development of varieties that are tolerant to water deficiencies. Plants have adapted to employ a variety of strategies to survive in water-scarce environments, generally leveraging one or more of three fundamental phenomena: escape, tolerance, and resistance mechanisms [6]. Escape mechanisms enable plants to finish their life cycle before drought sets in, successfully escaping unfavourable conditions. On the other side, tolerance mechanisms include physiological changes that allow the plant to deal with drought situations, such as stomatal closure to lower transpiration rates [7]. Finally, drought stress resistance mechanisms enable plants to actively combat its effects by increasing photosynthetic pigmentation and boosting the root-to-shoot ratio for effective assimilate partitioning. We may pave the way for the growth of crop types that remain strong in the face of drought stress by learning more about these systems and the traits shown by plants in water-deficient conditions [8].

Drought stress tolerance is a complex trait, governed by polygenic controls that exhibit low heritability and are significantly influenced by genotype × environment interactions. This complexity is further exacerbated when drought conditions coincide with other biotic and abiotic stresses, creating a multifaceted challenge for plant resilience and agricultural sustainability [9]. In this demanding context, the exploitation of advanced genomic tools appears to be a powerful strategy to promote the resilience of bread wheat to drought. Genome-wide association studies (GWAS) are at the forefront of this effort, providing a powerful approach to identifying genetic markers associated with drought tolerance. By dissecting the complex genetic architecture underlying drought tolerance, GWAS facilitates the identification of markers trait association (MTAs) and quantitative trait loci (QTLs), as well as candidate genes that could be exploited to develop drought-tolerant wheat varieties [3,10]. Identifying genomic regions associated with drought tolerance through GWAS and QTL mapping has become a key strategy in developing drought-resilient wheat varieties [11,12]. These methods allow for the identification of markers and QTLs that can be used in marker-assisted breeding programs to improve both yield and stress-related traits [13]. Several studies have mapped QTLs associated with important agronomic traits, including grain yield and drought susceptibility indices, under stress conditions. For example, Tahmasebi et al. [14] and Sobhanian et al. [15] identified QTLs for yield components and physiological traits in wheat populations exposed to terminal drought and heat stress, highlighting the genetic complexity of drought tolerance mechanisms. Similarly, Salarpour et al. [16] mapped QTLs for drought tolerance indices in doubled haploid wheat lines, providing critical insights into breeding values under varying environmental conditions.

Genomic prediction, which utilizes the information of genome-wide markers to predict the phenotypic performance of untested genotypes, offers a promising pathway to accelerate the breeding of drought-resistant wheat varieties. This approach not only expedites the breeding process but also enhances the precision in selecting genotypes with superior drought tolerance attributes [17].

In line with fundamental mechanisms of drought adaptation, our study aims to identify genotypes that exhibit enhanced drought tolerance by investigating grain yield performance and drought tolerance indices under both rain-fed and irrigated conditions. By evaluating elite bread wheat genotypes through GWAS and genomic prediction, we seek to pinpoint specific markers associated with drought tolerance. The insights into the plant's mechanisms for adapting to drought provide a framework for interpreting the observed variations in drought indices and grain yield, aligning with our objective to advance the development of drought-tolerant wheat varieties. The overall aim of this study was to identify the most reliable proxy for both drought tolerance and grain yield. In particular, the current study was carried out to 1) Evaluate the phenotypic variability of 198 elite bread wheat genotypes sourced from the International Center for Agricultural Research in the Dry Areas for drought stress tolerance under both irrigated and rain-fed conditions. 2) Employ GWAS to identify significant MTAs for grain yield and seven drought indices. 3) Estimate the prediction accuracy of the assessed indices using the genomic best linear unbiased prediction (gBLUP) model.

2. Material and methods

2.1. Plant material and experimental design

For this study, a total of 198 bread wheat genotypes (Supplementary Table 1). The genotypes used in this study were a collection of drought-tolerant bread wheat lines developed by the International Center for Agricultural Research in the Dry Areas (ICARDA). These lines were specifically bred for their resilience to drought and other environmental stresses, making them ideal candidates for evaluating grain yield and drought tolerance indices under both irrigated and rain-fed conditions. The lines were selected based on their genetic diversity and potential for improving drought tolerance in wheat breeding programs. These breeding lines were tested over two crop seasons (2021 and 2022) at the Sidi Al-Aidi station in Morocco (located at 33°07'27.600“ N, 7°37'43.800” W, at an altitude of 406 m above sea level). The study was conducted both under rain-fed and irrigated conditions, with an average precipitation of about 150 mm over the two years in the Sidi Al-Aidi region. For the rain-fed trials, no supplemental irrigation was provided, and the crops were watered solely by natural precipitation. In contrast, the irrigated trials received regular watering twice per week, whenever soil moisture levels dropped below field capacity, ensuring optimal growth conditions. The genotypes were planted across three m^2 plots following the alpha lattice design, with two replications. The soil at the Sidi Al-Aidi station is of the vertisol type, and the area has a moderate humidity atmosphere with temperatures ranging annually from 10 to 40 °C. Planting was carried out in the first week of December at a rate of 100 kg per hectare.

2.2. Drought indices analysis

The grain yield (GY) performance of genotypes was evaluated under both irrigated and rain-fed conditions. Initially measured in kilograms, the yields were subsequently converted to tons per hectare (t/ha) for standardized reporting. Various indices were determined to evaluate the performance of the genotypes under drought conditions; these included the stress tolerance index (STI), drought susceptibility index (DSI), tolerance index (TOL), yield stability index (YSI), mean productivity (MP), and geometric mean productivity (GMP). To derive these indices,

Table 1
Formulas (equation) of drought indices.

Drought indices	Equation	References
Stress Tolerance Index (STI)	$STI = \frac{(Yp \times Ys)}{(\overline{Y}p^{2})}$	[18]
Drought Susceptibility Index (DSI)	$DSI = \frac{1-\left(\frac{Ys}{Yp}\right)}{1-\left(\frac{\overline{Y}s}{\overline{Y}p}\right)}$	[19]
Tolerance Index (TOL)	$TOL = Yp - Ys$	[20]
Yield Stability Index (YSI)	$YSI = \frac{Ys}{Yp}$	[21]
Mean Productivity (MP)	$MP = \frac{(Yp + Ys)}{2}$	[18]
Geometric Mean Productivity (GMP)	$GMP = \sqrt{Yp \times Ys}$	[18]

$\boldsymbol{Yp}$: the yield under irrigated conditions, $\boldsymbol{Ys}$: the yield under drought conditions, $\overline{\boldsymbol{Y}}\boldsymbol{p}$: the mean yield under irrigated conditions, $\overline{\boldsymbol{Y}}\boldsymbol{s}$: the mean yield under stress (drought) conditions.

the grain yield data from both irrigated and rain-fed conditions were utilized for each plot. These indices were calculated from the grain yield performance of genotypes evaluated under the two growth conditions (Table 1).

2.3. Genotyping

The wheat samples were genotyped using the Wheat Illumina iSelect 15K single nucleotide polymorphism (SNP) array at the SGS Institut Fresenius GmbH TraitGenetics Section located in Gatersleben, Germany. Samples were collected from the flag leaves of the individual genotypes, placed in plates, and then sent to the institute for DNA extraction and genotyping. In total, 198 bread wheat genotypes were analyzed using 15K SNP markers. Following the data collection, the genotypic data underwent a filtering process to retain only the most reliable data for analysis. Specifically, SNPs with a minor allele frequency (MAF) below 5 % and heterozygosity exceeding 10 % were excluded. After this filtration process, 13,151 SNPs remained, forming the basis for subsequent GWAS and genomic prediction analyses.

2.4. Statistical analyses, linkage disequilibrium and population structure

Statistical analysis of the phenotypic data was conducted across schemes and years following linear mixed model:

$$y_{ij} = \mu + G_i + E_j + GE_{ij} + \varepsilon_{ij}$$

Where y_{ij} denotes the observed phenotypic value of the i-th genotypes in the j-th environemet, μ is the common intercept, G_i represents the effect of the i-the genotype, E_j is the effect of the j-th environment, GE_{ij} captures the interaction effect between the i-th genotype and the j-th environment, and εij accounts for the residual error in the model for the observed phenotypic value. Except for μ, all effects were treated as random to estimate the variance components and broad sense heritability. While genotypes were considered fixed effects to derive the Best Linear Unbiased Estimations (BLUEs).

The descriptive statistics including the mean, maximum, minimum, standard deviation, and coefficient of variation were computed using the "RcmdrMisc" package in R. Linkage disequilibrium (LD) was assessed using the Tassel software, wherein the LD values for the 198 bread wheat genotypes were analyzed based on the SNP markers distributed across the wheat genomes. The methodology for the LD decay investigation was derived from a protocol established by Remington and colleagues [22]. Additionally, a principal component analysis (PCA) was employed to elucidate the population structure.

2.5. Genome-Wide Association study and genes annotation

A genome-wide association study (GWAS) focusing on grain yield and drought indices was executed utilizing the Genomic Association and Prediction Integrated Tool version 3 (GAPIT 3) in R language, as documented by Ref. [23]. Within this study, we adopted the mixed linear model (MLM), incorporating principal components (PCs) and a kinship K matrix in the formula: phenotype = marker + PCs + kinship + error. Significant markers were visually represented through Manhattan plots, identifying markers with a -Log10(p) value exceeding 3.0 as substantial marker-trait associations (MTAs) dispersed across all 21 chromosomes. To control for false positives in our GWAS analysis, we applied the False Discovery Rate (FDR) correction across the identified markers, adjusting the p-values to account for multiple testing. We used a -log10(p) threshold of 3 for significance, which is consistent with similar studies. However, we acknowledge that this threshold may be considered relatively low given the marker density. The CMplot package in the R environment facilitated the depiction of markers on Manhattan and QQ plots, adhering to a threshold of -Log10(p) > 3, following guidance by Ref. [24]. Subsequently, genes associated with the significant markers were pinpointed utilizing the EnsemblPlants platform, leveraging the Variant Effect Predictor function to spotlight functional genes. In the gene annotation process of our study, the reference genome used was the IWGSC RefSeq v1.0. This genome served as the basis for all genomic alignments and annotations. Marker-gene associations were determined based on proximity within the genome, with significant markers identified through GWAS being aligned to the nearest gene loci within a predefined window. This approach allowed for the identification of candidate genes potentially associated with the traits under investigation. Specific parameters, including the maximum allowable distance from the marker to the gene and the annotation pipeline details, were employed as per standard practices in genomic studies of Triticum aestivum, ensuring accurate and relevant gene annotations. Further analysis was conducted on the UniProt platform to delineate the proteins encoded by these genes, elaborating on the molecular functions and biological processes spearheaded by these proteins. The SNPs epistatic interactions were analyzed based on the effects of significant markers, utilizing the following formulas:

- The multiplicative combined effect:

$$\boldsymbol{Effect}_{combined\_multiplicative} = \boldsymbol{Effect}_{SNP1} \times \boldsymbol{Effect}_{SNP2} \quad (1)$$

- The interaction strength for the multiplicative model

$$\boldsymbol{Interaction\ Strength}_{multiplicative} = \boldsymbol{Effect}_{combined\_multiplicative} - (\boldsymbol{Effect}_{SNP1} + \boldsymbol{Effect}_{SNP2}) \quad (2)$$

The Circos representation of SNP epistasis was generated using ClicO FS software [25], where only SNPs demonstrating high synergy were tracked and displayed.

2.6. Genomic prediction

To conduct genomic prediction (GP) of all the drought indices and grain yield, we employed the genomic best linear unbiased prediction (gBLUP) model using TASSEL version 5.2.70, as cited by Ref. [26]. Where 50 genotypes were used as breeding population and 148 genotypes were used as training population. The accuracy of the genomic prediction was evaluated through a five-fold cross-validation implemented over 20 iterations within the gBLUP model-based TASSEL framework.

3. Results

3.1. Descriptive statistics and lattice ANOVA

Fig. 1 shows the unimodal distribution of grain yields under both irrigated and rain-fed conditions, as well as the unimodal distribution of all drought indices, across the set of 198 bread wheat genotypes. The statistical analyses of the drought indices for 198 bread wheat genotypes are detailed in Table 2. Grain yield under irrigated conditions fluctuated between 1.12 and 4.83 t/ha, with a mean value of 2.87 t/ha and a coefficient of variance (CV) of 23.25 %. In comparison, the yield under rain-fed conditions ranged from 0.34 to 2.57 t/ha, averaging 1.22 t/ha with a CV of 35.4 %. The STI index showed a promising sign of good drought tolerance with values stretching from 0.06 to 1.17 and a mean of 0.44. Meanwhile, the DSI index indicated better drought tolerance with figures extending from −0.69 to 0.58 and a central tendency of 0.14. The TOL index demonstrated the most considerable variation with a CV of 45.21 %, harboring a mean of 1.63 within a scope of −0.41 to 3.81. In contrast, the MP index depicted the lowest fluctuation, characterized by a mean of 2.01 within a 0.93 to 3.12 range and a minimal CV of 22.2 %. Other indices such as the YSI and GMP indexes bore mean values of 0.44 and 1.8 with CVs of 43.1 % and 24.2 %, respectively. Generally, the genotypes manifested considerable drought tolerance, with genotypes G82 (HUBARA-13//ACHTAR/INRA 1764) and G150 (TEMPORALERA M

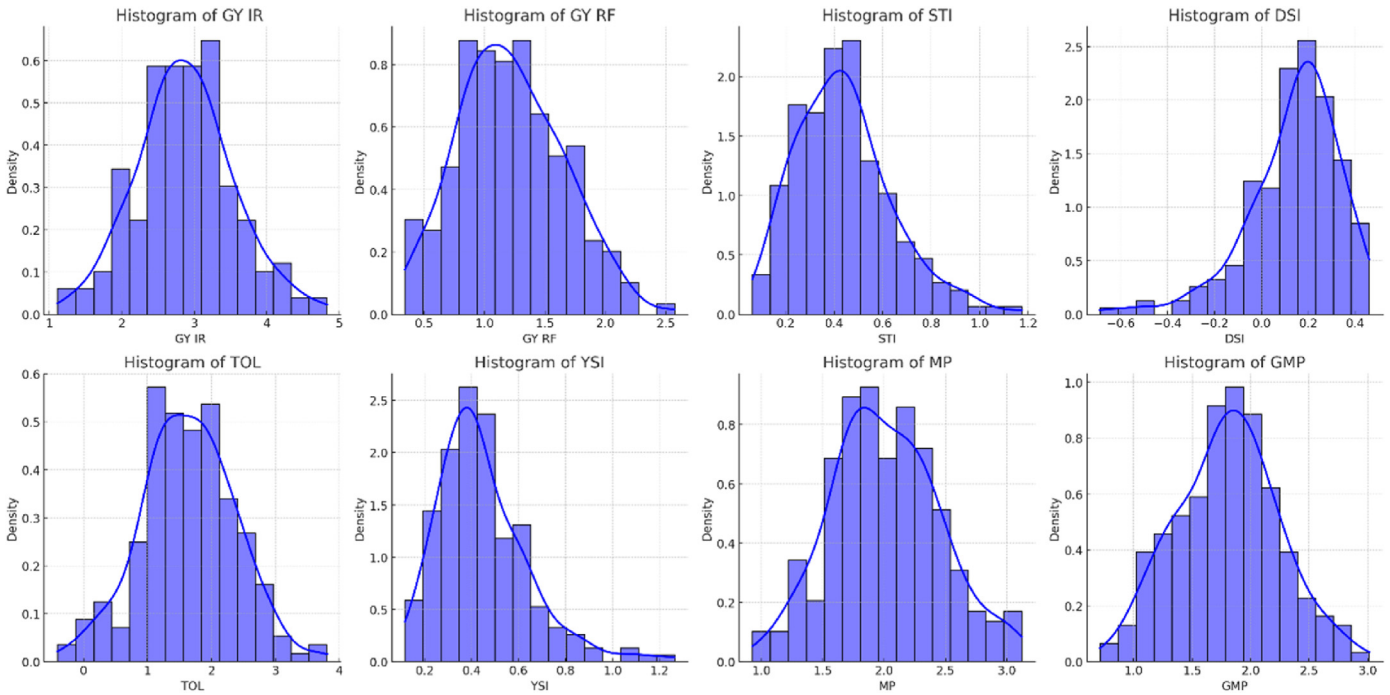

Fig. 1. Distribution of genotypes performance for grain and drought indices of 198 bread wheat genotypes at Sidi Al-Aidi station.

Table 2
Statistical description of 198 bread wheat genotypes for grain yield and drought indices.

	Mean	Min	Max	Sd	Se	CV (%)	H^2
GY IR (t/ha)	2.87	1.12	4.83	0.67	0.05	23	0.73
GY RF (t/ha)	1.42	0.95	2.57	0.33	0.02	23.2	0.53
STI	0.51	0.29	1.17	0.16	0.01	32.37	0.57
DSI	0.14	−0.69	0.58	0.20	0.01	–	0.60
TOL	1.81	1.01	3.04	0.51	0.04	28	0.60
YSI	0.50	0.30	1.26	0.17	0.01	34.64	0.59
MP	2.01	0.93	3.12	0.45	0.03	22.22	0.63
GMP	1.80	0.71	3.01	0.43	0.03	24.20	0.56

SD: standard deviation; SE: standard error; H^2: heritability; CV: coefficient of variation; GY_IRR: grain yield under irrigated conditions; GY_RF: grain yield under rain-fed conditions; STI: Stress Tolerance Index, DSI: Drought Susceptibility Index; TOL: Tolerance Index; YSI: Yield Stability Index; MP: Mean Productivity; GMP: Geometric Mean Productivity.

87*2/KONK//FAYEQ-1) recording the pinnacle of grain yield under rain-fed conditions, documenting yields of 4.83 and 4.72 t/ha, respectively. Furthermore, G160 recorded a notable STI index of 1.17, with G59 not far behind with an STI value of 1.04.

The lattice ANOVA result (Table 3) presents the analysis of variance for grain yield (GY) and various drought indices across 198 bread wheat genotypes. The variation is divided into three main factors: Genotype, Replication, and Block. For each source of variation, the p values are provided for grain yield under irrigated (GY IR) and rainfed (GY RF) as well as drought tolerance indices. The Genotype effect shows significant p-values for most traits, suggesting a strong genetic influence on these traits. Low p-values recorded for GY RF under the Genotype category highlight low genetic variability among the genotypes for these traits. Most of the indices showed significant variances with genotype as a factor. In contrast, the Replication and Block effects show generally lower mean square values, indicating the consistency of the genotypes' performance across replications and blocks. The presence of significant values in the Replication and Block rows, like for GY IR and TOL under Replication and Block, suggests some environmental or experimental variation. In the lattice ANOVA table (Table 3), the source labeled 'Block' refers to the blocking factor used in our experimental design. Blocking was implemented to control for variability due to environmental gradients across the experimental field. Specifically, the experiment was divided into 26 blocks, each containing a representation of all treatments. This design allows us to minimize the impact of soil, microclimate, and other local variations, thus providing a more reliable assessment of the effects due to genotypes and replication. The p-values listed under 'Block' indicate the statistical significance of block effects for each trait measured, helping to discern the intrinsic genetic differences from those influenced by environmental variations.

3.2. *Principal component analyses (PCA) and correlation*

The current panel showed three clearly clustered groups (Fig. 2A). The third group comprised the highest number, with 93 genotypes included, followed by group 2 with 63 and the remaining 44 genotypes clustered in group 1. The biplot analysis with the first two PCs elucidates the association between grain yield and various drought indices (Fig. 2B). The first and second principal components accounted for 52.6 % and 41.7 % of the total phenotypic variation recorded from grain yield and drought indices, respectively. The PCA biplot illustrates the relationships

Table 3
Lattice analysis of variance (ANOVA) of grain yield under both irrigated and drought conditions, as well as for drought tolerance indices, using mean square values.

Sources of variance	Df	GY IR	GY RF	STI	DSI	TOL	YSI	MP	GMP
Genotype	197	0.86 ***	0.36	0.07 **	0.07 **	1.04 ***	0.07 **	0.35 ***	0.37 **
Replication	1	1.78 *	0.009	0.05	0.07	2.05 *	0.07	0.38	0.05
Block	26	0.39	0.47	0.07	0.08	0.82 *	0.08 *	0.22	0.37

*, **, *** significant at 0.05, 0.01 and 0.001 levels, respectively.

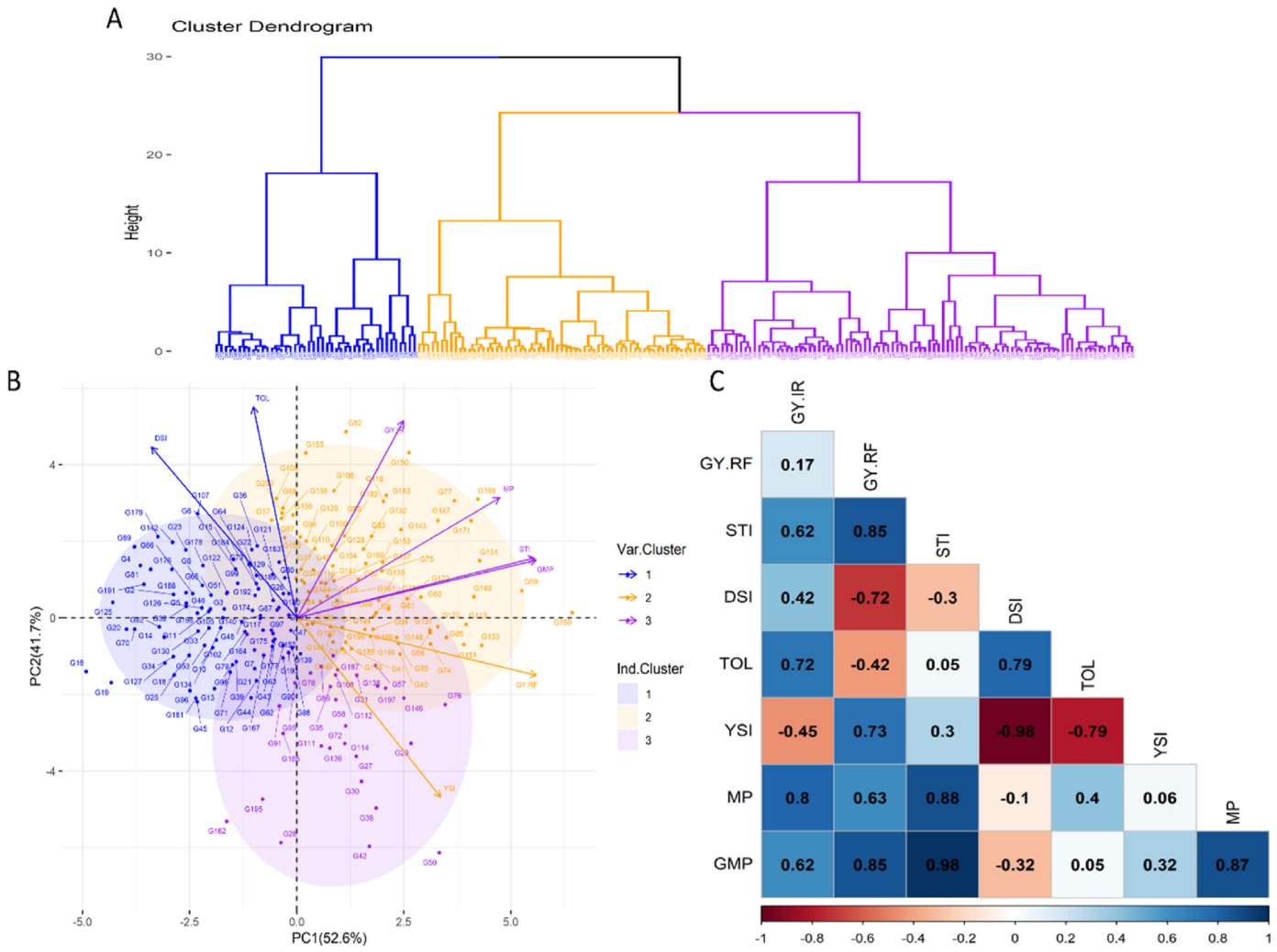

Fig. 2. The cluster dendrogram (A) illustrates the relationships among 198 bread wheat genotypes; PCA (B) visually represents the principal component analysis results; and the correlation graph (C) demonstrates the relationships between grain yield and drought indices across the 198 bread wheat genotypes.

between the traits by showing the angles between their vectors. Traits with smaller angles between their vectors are more positively correlated, while traits with larger angles, closer to 180°, indicate negative correlations. For instance, the vectors for GY_IR and GMP form a small angle, suggesting a strong positive correlation between these traits. In contrast, DSI forms a larger angle with YSI, indicating a negative correlation, which is expected since higher drought susceptibility would result in lower yield stability. Fig. 2C shows the correlations among drought indices. STI and GMP exhibited a highly significant positive correlation (R = 0.98) followed by STI and MP (R = 0.88). GMP also held a significant positive correlation with MP (R = 0.87) and with GY-RF and GY-IR, with correlation coefficients of 0.85 and 0.62, respectively. Conversely, YSI had a significant negative correlation with DSI, TOL, and GY-IR, correlation coefficients −0.98, −0.79, and −0.45, respectively. Additionally, grain yield under rain-fed conditions was negatively correlated with DSI and TOL with-0.72 and −0.42, respectively.

3.3. Linkage disequilibrium (LD) and neighbor-joining (NJ) tree

The Linkage Disequilibrium (LD) decay exhibited distinct patterns across the A, B, and D genomes. For genome A, a total of 294,726 SNP marker pairs were evaluated. The average R2 was 0.17, and the LD decayed at a distance of 3.34 Mbp (Fig. 3A). A total of 348,876 pairs of SNP marker comparison was done for genome B, and the average R2 value was 0.18 while LD decayed at 4.52 Mbp (Fig. 3B). A relatively few SNP markers were identified from the D genome, resulting in only 135,176 pair-wise SNP markers comparison, and the mean LD value (R2) was 0.15, and LD decayed at 3.88 Mbp (Fig. 3C). Fig. 3D showcases the Neighbor-Joining (NJ) tree constructed from the genotypic data of 198 bread wheat genotypes. This NJ tree delineates three distinct groups, each highlighted in a different color.

3.4. Genome wide association study and candidate genes

In this study, we identified a total of 39 significant markers ($p < 0.001$) and 23 QTLs associated with several variables, including grain yield under both irrigated and rain-fed conditions, along with DSI, GMP, MP, STI, TOL, and YSI indices. These findings, documented at the Sidi Al-Aidi station, are detailed in Tables 3 and 4, and illustrated in Figs. 4 and 5. Fig. 6 shows SNPs density on each wheat chromosome of 198 bread wheat, genotypes (see Tables 4 and 5).

Table 3 delineates that Marker Trait Associations (MTAs) correlated with Quantitative Trait Loci (QTLs) of various indices and grain yield under different conditions.

Assessing the grain yield under irrigated (GY.IR) conditions revealed 2 salient MTAs and 2 QTLs dispersed across chromosomes 2A and 5A. The marker "*BobWhite_c42349_99*" recorded a -log10(p) score of 3.31, localized on chromosome 5A at the 665.47 Mbp position, this marker is linked to the QTL "*QTL_ISBW_GY.1*". Furthermore, the marker "*wsnp_Ex_c11827_18986376*" was recorded for GY.IR. This marker was

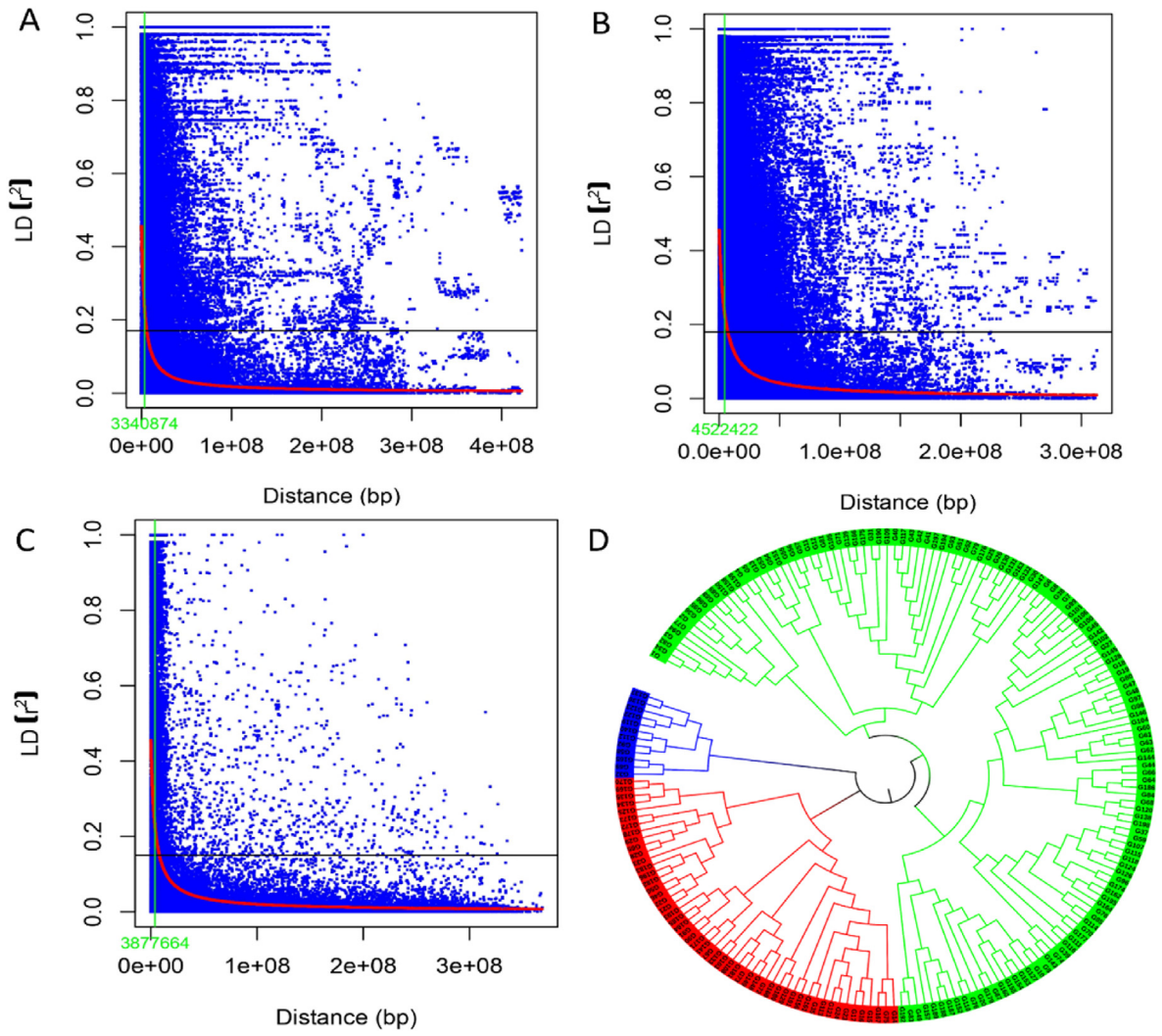

Fig. 3. Illustration of the linkage disequilibrium decay in genome A (A), genome B (B), and genome D (C), alongside the Neighbor-Joining (NJ) tree (D) constructed from the genotypic data of 198 bread wheat genotypes.

Table 4
List of the significant markers and QTLs associated with GY.IR, GY.RF, DSI, and GMP indices and grain yield under irrigated and rain-fed conditions from GWAS of 198 bread wheat genotypes at Sidi Al-Aidi station.

Trait	Marker	QTL	Chro	Pos	Effect	-Log10(p)
GY.IR	*BobWhite_c42349_99*	*QTL_ISBW_GY.1*	5A	665471358	−0.42	3.31
	wsnp_Ex_c11827_18986376	*QTL_ISBW_GY.2*	2A	733922181	−0.37	3.15
GY.RF	*wsnp_Ex_rep_c66358_64543089*	-	UN	300833704	0.23	3.20
	wsnp_Ex_c12127_19394952	-	5B	418010902	0.92	3.14
	Kukri_rep_c104611_210	*QTL_ISBW_GY.3*	2A	709850045	−0.23	3.07
	wsnp_Ra_c10658_17500498	*QTL_ISBW_GY.4*	2B	689871439	0.23	3.03
	Kukri_c51101_351	*QTL_ISBW_GY.5*	7B	630107823	0.90	3.03
DSI	*BS00049927_51*	-	1B	108837029	−0.31	3.66
	wsnp_Ex_c214_421541	-	5B	42947782	−0.21	3.58
	AX-94951542	-	5B	42393918	−0.12	3.10
	CAP11_c1087_327	*QTL_ISBW_DSI.1*	6B	3883815	0.15	3.42
	wsnp_Ex_c31672_40435001	-	5A	631668358	−0.29	3.39
	wsnp_Ex_c3834_6971470	-	5B	536516528	−0.39	3.34
	BS00050522_51	*QTL_ISBW_DSI.2*	1B	1433694	−0.57	3.21
	BobWhite_c42349_99	*QTL_ISBW_DSI.3*	5A	665471358	−0.12	3.18
GMP	*Kukri_c51101_351*	*QTL_ISBW_GMP.1*	7B	630107823	0.92	3.43
	AX-94430599	*QTL_ISBW_GMP.2*	2D	601601896	0.25	3.26
	wsnp_Ex_c12127_19394952	-	5B	418010902	0.64	3.24
	wsnp_Ra_rep_c69620_67130107	*QTL_ISBW_GMP.3*	7A	85612705	0.55	3.06
	AX-94533562	*QTL_ISBW_GMP.3*	7A	85912049	0.27	3.03

QTL: Quantitative Trait Loci; Chro: Chromosome; Pos: Position.

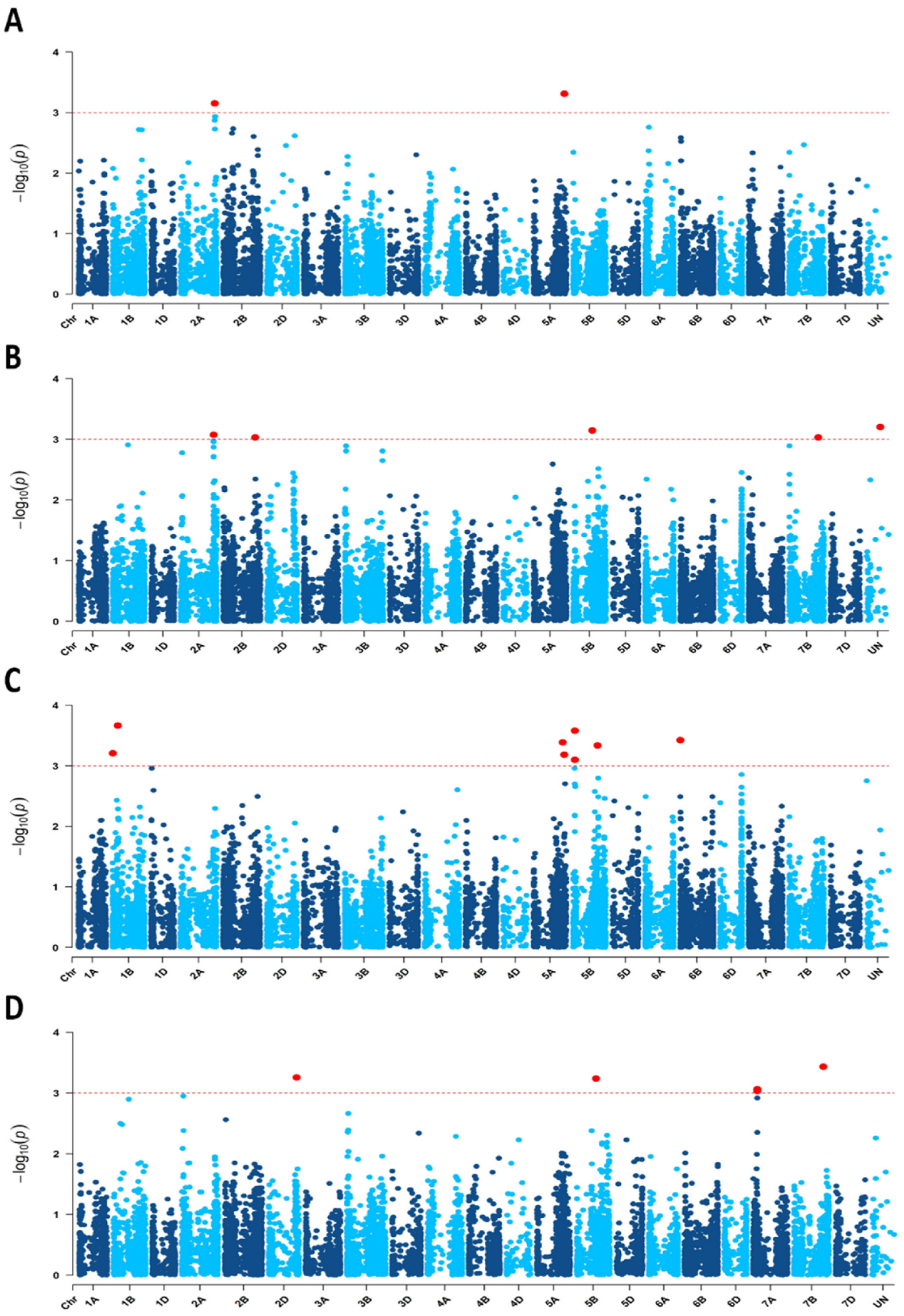

Fig. 4. Manhattan and QQ plots for 198 bread wheat genotypes evaluating grain yield under both irrigated (A) and rain-fed (B) conditions, as well as DSI (C), and GMP (D) indices, as recorded at the Sidi Al-Aidi station.

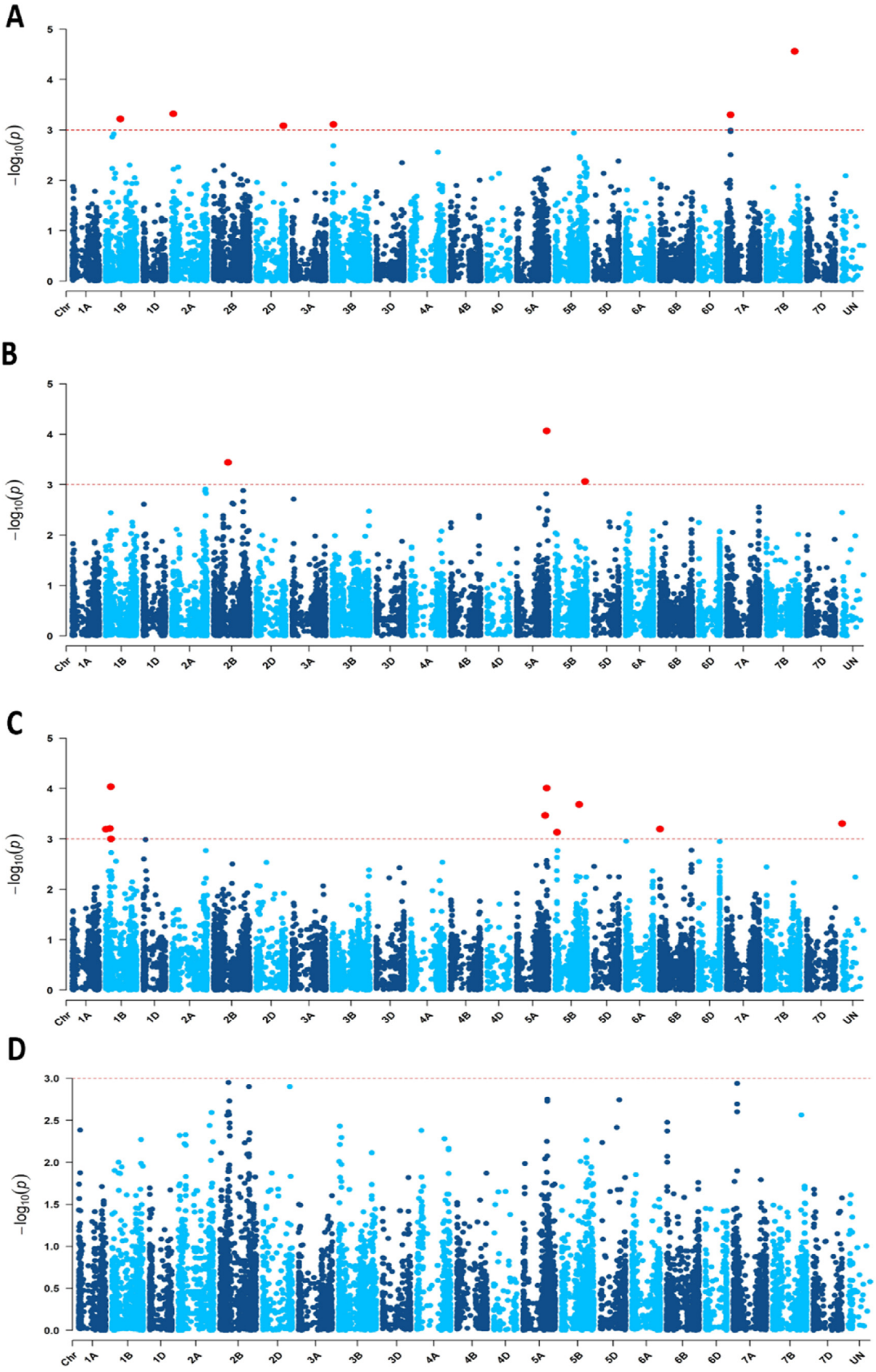

Fig. 5. Manhattan and QQ plots for 198 bread wheat genotypes evaluating STI (A), TOL (B), YSI (C), MP (D) indices, as recorded at the Sidi Al-Aidi station.

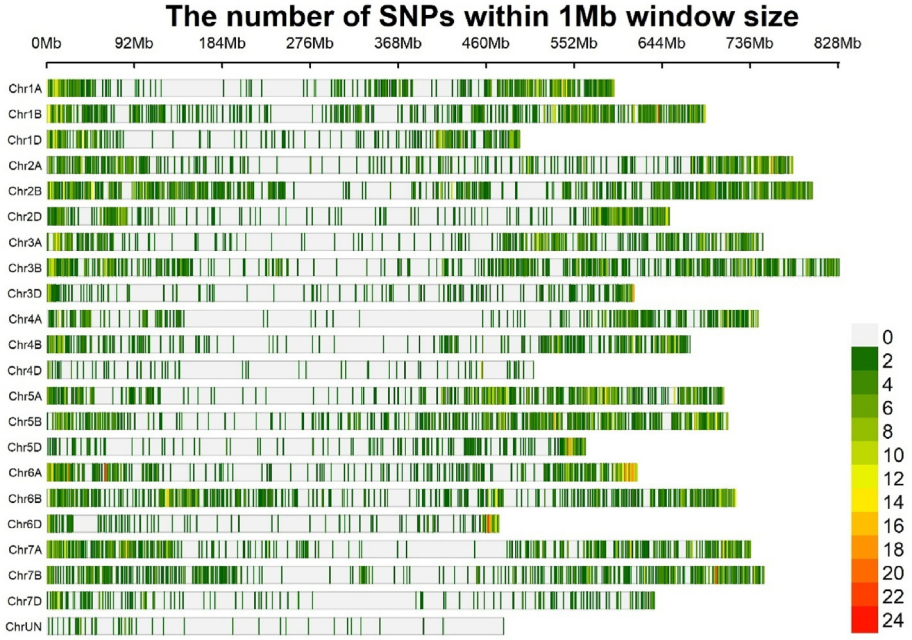

Fig. 6. SNPs density on each wheat chromosome of 198 bread wheat genotypes.

Table 5
List of the significant marker and QTLs associated with STI, TOL and YSI indices from GWAS of 198 bread wheat genotypes at Sidi Al-Aidi station.

Trait	Marker	QTL	Chro	Pos	Effect	-Log10(p)
STI	*Kukri_c51101_351*	*QTL_ISBW_STI.1*	7B	630107823	0.51	4.56
	AX-94381659	*QTL_ISBW_STI.2*	2A	29871296	0.16	3.32
	AX-94533562	*QTL_ISBW_STI.3*	7A	85912049	0.11	3.30
	AX-94467830	-	1B	323618700	0.13	3.22
	wsnp_Ex_c57450_59156677	*QTL_ISBW_STI.4*	3B	24944078	0.34	3.11
	AX-94430599	*QTL_ISBW_STI.5*	2D	601601896	0.11	3.08
TOL	*BobWhite_c42349_99*	*QTL_ISBW_TOL.1*	5A	665471358	−0.52	4.07
	AX-95009146	-	2B	315664134	−0.71	3.44
	BS00078784_51	*QTL_ISBW_TOL.2*	5B	668198232	−1.43	3.06
YSI	*Ra_c455_283*	-	1B	117180060	0.09	3.00
	BS00049927_51	-	1B	108837029	0.30	4.04
	BobWhite_c42349_99	*QTL_ISBW_YSI.1*	5A	665471358	0.13	4.01
	wsnp_Ex_c3834_6971470	-	5B	536516528	0.37	3.68
	wsnp_Ex_c31672_40435001	-	5A	631668358	0.28	3.47
	BS00023006_51	-	UN	6017714	0.15	3.31
	BS00084305_51	*QTL_ISBW_YSI.2*	1B	90992756	0.19	3.21
	CAP11_c1087_327	*QTL_ISBW_YSI.3*	6B	3883815	−0.14	3.19
	BS00050522_51	*QTL_ISBW_YSI.4*	1B	1433694	0.55	3.19
	wsnp_Ex_c214_421541	-	5B	42947782	0.19	3.13

QTL: Quantitative Trait Loci; Chro: Chromosome; Pos: Position.

located on chromosome 2A at position 733.92 Mbp. Grain yield under rain-fed conditions recorded 5 MTAs and 3 QTLs, predominantly on chromosomes 2A, 2B, 5B, and 7B. The highest marker here was "*wsnp_Ex_rep_c66358_64543089*", recorded a highest -log10(p) figure of 3.20 on unknown chromosome at position 300.83Mbp, followed by "*wsnp_Ex_c12127_19394952*" with a -log10(p) = 3.14 positioned on chromosome 5B at the position 418.01 Mbp.

The Drought Susceptibility Index (DSI) recorded 8 significant MTAs and 3 QTLs, with the marker "*BS00049927_51*" was the most significant at -log10(p) = 3.66; it is situated on chromosome 1B at a position of 108.83 Mbp. Furthermore, markers "*wsnp_Ex_c214_421541*", "*AX-94951542*" and "*wsnp_Ex_c3834_6971470*", found on chromosome 5B, had respective -log10(p) values of 3.58 and 3.10, and 3.34 and were located at positions 42.94 Mbp, 42.39 Mbp, and 536.51 Mbp. The Geometric Mean Productivity (GMP) index highlighted 5 significant MTAs and 4 QTLs; the highest was the "*Kukri_c51101_351*" marker with a -log10(p) value of 3.43, identified on chromosome 7B at position 630.10 Mbp and incorporated within the "*QTL_ISBW_GMP.1*" QTL, followed by the "*AX-94430599*" marker which holds a -log10(p) value of 3.26 located on chromosome 2D at position 601.60 Mbp. Another notable marker in this context was "*AX-94533562*," which was aligned with the GMP index and recorded the lowest log10(p) value of 3.03, located on chromosome 7A at position 85.9 Mbp.

Table 4 shows the significant MTAs correlated with QTLs of STI, TOL and YSI indices. The STI index recorded 6 significant MTAs and 5 QTLs, with the marker "*Kukri_c51101_351*" recorded as the most significant marker at -log10(p) = 4.56; it is situated on chromosome 7B at a position of 630.10 Mbp. Furthermore, markers "*AX-94381659*"and "*AX-94533562*"found on chromosome 2A and 7A, respectively, had respective -log10(p) values of 3.32 and 3.30 and were located at positions 29.87 Mbp, 85.91 Mbp. The TOL index recorded 3 significant MTAs and 2 QTLs, the highest marker "*BobWhite_c42349_99*" recorded a -log10(p) value of 4.07, identified on chromosome 5A at position 665.47 Mbp, this marker was alongside with the marker "*AX-95009146*" located on chromosome

2B at position 315.66 Mbp. The marker "*BS00078784_51*" on QTL "*QTL_ISBW_TOL.2*" recorded the lowest -log10(p) = 3.06 located on the chromosome 5B at position 668.19 Mbp. The Mean YSI index recorded 10 significant MTAs and 4 QTLs, the marker "*BS00049927_51*" recorded the highest -log10(p) = 4.04 located on the chromosome 1B at position 108.83 Mbp, whereas the marker "*Ra_c455_283"* recorded the lowest -log10(p) = 3 located on chromosome 1B at position 117.18 Mbp.

Fig. 7 illustrates the SNPs characterized by the highest degrees of synergy epistasis. In particular, the SNPs "*BobWhite_rep_c66990_294*" and "*RAC875_c583_391"* showcase the pinnacle of synergy interaction, registering the most substantial strength of the multiplicative interaction effect at 2.239. This is closely trailed by the synergy between markers "*wsnp_BE498786B_Ta_2_1*" and "RAC875_c583_391," which exhibits a strength of multiplicative interaction effect amounting to 2.155. Contrarily, the least synergic interaction was noted between the markers "*Excalibur_rep_c107577_250*" and "*BS00062731_51*," reflecting a minimal strength of multiplicative interaction effect of 0.00012. Notably, the pair consisting of markers "*BS00070104_51*" and "*RAC875_c583_391*" demonstrated the highest antagonistic SNP epistasis, with a strength of multiplicative interaction effect at −1.087.

3.5. Gene annotation

In this study, 14 candidate genes were identified and correlated with grain yield under both irrigated and rain-fed conditions alongside various drought indices (Table 6). The table provided appears to be an excerpt from a gene annotation report associated with the GWAS analyses. The table lists various SNP markers and associates them with specific genes, their protein products, molecular functions, and biological processes. For instance, the marker *BS00050522_51* is linked to the gene *TraesCS1B02G001800*, which encodes for the protein Phospholipid-transporting ATPase. This protein is involved in ATPase-coupled intramembrane lipid transporter activity, playing a role in the biological process of phospholipid translocation. Similarly, *AX-94381659* is associated with the 40S ribosomal protein S6, which is essential for protein synthesis as part of the ribosomal structure and function in the process of translation. The marker "*wsnp_Ex_c57450_59156677*" was linked to the gene TraesCS3B02G049100 which encodes for Protein kinase domain-containing protein involved in protein kinase activity and phophorylation. The genes that we found in this study encode for Metal tolerance protein 1-like, RRM domain-containing protein, Catalase, NusG-like N-terminal domain-containing protein, and Thioredoxin domain-

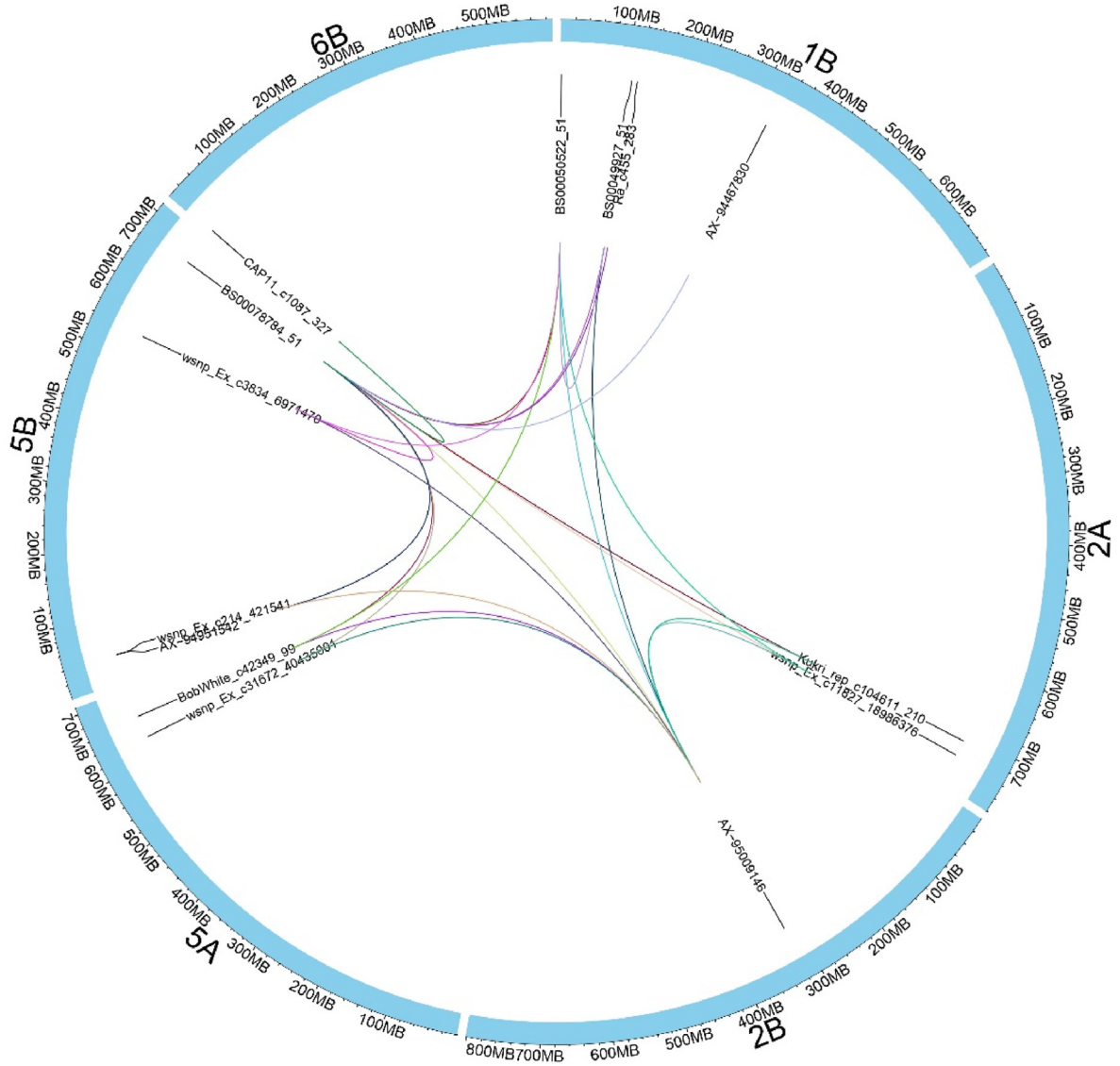

Fig. 7. SNPs are characterized by the highest degrees of synergy epistasis.

Table 6
List of putative genes associated with significant markers.

Marker	Gene	Protein	Molecular function	Biological prosess	Reference
BS00050522_51	*TraesCS1B02G001800*	Phospholipid-transporting ATPase	ATPase-coupled intramembrane lipid transporter activity	Phospholipid translocation	[27]
BS00084305_51	*TraesCS1B02G090400*	Metal tolerance protein 1-like	Zinc ion transmembrane transporter activity	Zinc ion transmembrane transport	[28]
AX-94381659	*TraesCS2A02G066100*	40S ribosomal protein S6	Structural constituent of ribosome	Translation	[29]
Kukri_rep_c104611_210	*TraesCS2A02G464300*	RRM domain-containing protein	RNA binding	–	[30]
wsnp_Ex_c11827_18986376	*TraesCS2A02G505800*	NADPH–cytochrome P450 reductase	NADPH-hemoprotein reductase activity	–	[31]
wsnp_Ra_c10658_17500498	*TraesCS2B02G491600*	DUF4220 domain-containing protein	–	–	[32]
AX-94430599	*TraesCS2D02G508800*	STAS domain-containing protein	Monoatomic anion transmembrane transporter activity	–	[30]
wsnp_Ex_c57450_59156677	*TraesCS3B02G049100*	Protein kinase domain-containing protein	Protein kinase activity	Phosphorylation	[33]
BobWhite_c42349_99	*TraesCS5A02G498000*	Catalase	Catalase activity	Response to hydrogen peroxide	[34]
BS00078784_51	*TraesCS5B02G500900*	NusG-like N-terminal domain-containing protein	–	Transcription elongation-coupled chromatin remodeling	[35]
CAP11_c1087_327	*TraesCS6B02G005100*	Thioredoxin domain-containing protein	Protein disulfide isomerase activity	Protein folding	[36]
wsnp_Ra_rep_c69620_67130107	*TraesCS7A02G133400*	Transmembrane protein	–	–	[37]
AX-94533562	*TraesCS7A02G134100*	Galactinol–sucrose galactosyltransferase	–	Carbohydrate metabolic process	[38]
Kukri_c51101_351	*TraesCS7B02G366200*	Nudix hydrolase domain-containing protein	ADP-glucose pyrophosphohydrolase activity	Nucleoside phosphate metabolic process	[39]

containing protein; these proteins involved in zinc ion transmembrane transporter activity, catalase activity, response to hydrogen peroxide, and transcription elongation-coupled chromatin remodeling, respectively. Each entry in the table provides a detailed view of the potential function of the gene product and its role in the cell, allowing researchers to understand better the genetic basis of certain traits or conditions. This information is crucial for advancing our understanding of the biology of wheat under drought conditions.

3.6. Genomic prediction

The fivefold cross-validated predictions using the gBLUP model showed an average prediction accuracy between 0.31 and 0.50 for all drought indices and grain yield (Fig. 8). The analysis revealed that prediction accuracy was highest for the TOL index, where the highest value was 0.50. This was closely followed by the yield under irrigated conditions, with the highest value of accuracy was 0.43, and the DSI index, which had a score of 0.31. In a detailed breakdown of the drought indices, GMP recorded an accuracy of 0.34. MP and GMP have values of 0.33 and 0.34, respectively. Other indices recorded moderate accuracy levels; STI showcased a score of 0.38, while YSI and GY.RF recorded values of 0.41 and 0.40, respectively.

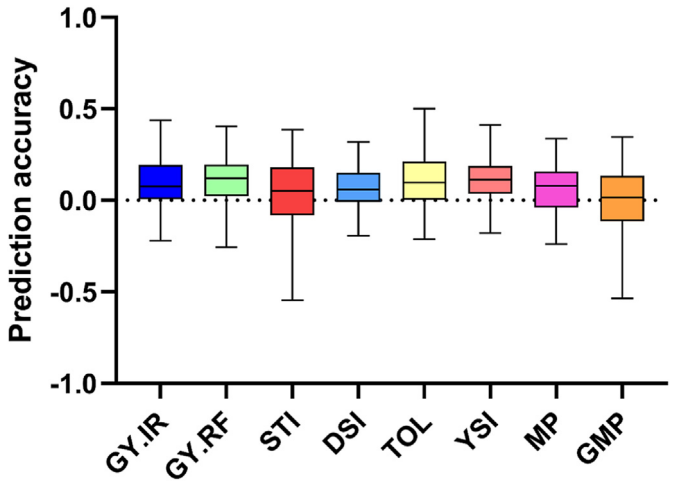

Fig. 8. Genomic prediction of grain yield under rain-fed and irrigated conditions, as well as drought indices.

4. Discussion

4.1. Phenotypic variability for grain yield and drought indices

Bread wheat (Triticum aestivum L.) occupies a central position as a primary food source in the Central and West Asia and North Africa (CWANA) and Sub-Saharan Africa (SSA) regions. These areas are consistently challenged by an array of abiotic and biotic stressors, notably drought, which emerges as the most significant. The CWANA region, in particular, experiences chronic and severe drought stress, markedly impairing wheat productivity and presenting a substantial constraint. In light of these exigent circumstances, it is imperative/urgan to develop the novel drought tolerance genotyes and cultivals used for the sustained cultivation of wheat in these regions [1]. The objective of ICARDA's breeding initiative for bread wheat is to explore genotypes that exhibit not only desirable end-use qualities but also resilience to both biotic and abiotic stressors. Grain yield is a complex trait underpinned by a multitude of genetic factors and profoundly influenced by the dynamic interplay between genotypic attributes and environmental variables [40]. In this investigation, we cultivated 198 distinct bread wheat genotypes developed by ICARDA under two disparate conditions: irrigated and rain-fed. These genotypes underwent rigorous assessment for drought tolerance, utilizing drought indices for each genotype as primary evaluative measures. These indices subsequently informed Genome-Wide Association Studies (GWAS) and genomic predictions, providing a comprehensive data corpus for analytical and predictive modelling. Contrary to prevailing assumptions that irrigation substantially enhances yield, our findings indicate that grain yield under rain-fed conditions did not significantly differ from that under irrigated scenarios. This observation challenges the conventional belief in the necessity of irrigation for optimal yield, positing a formidable potential for successful wheat cultivation in rain-fed contexts [41]. The approximate parity in yield between the two environments underscores the effectiveness of recent breeding strategies in augmenting drought tolerance, an advancement of paramount importance considering the global trends of fluctuating rainfall patterns and escalating water scarcity issues [42].

In this study, considerable variation was observed for yield and drought indices under rain-fed and irrigated schemes. We discovered promising indicators of drought resistance in the bread wheat genotypes we examined. Notably, the Stress Tolerance Index (STI) revealed a commendable average of 0.44, surpassing the mean STI value of 0.53 noted in previous research by Poudel et al. [43]. A particularly notable finding is the highest recorded STI value of 1.17 in one of our genotypes, demonstrating its potential for excellent performance under both stress-free and stressful conditions. Regarding the Drought Susceptibility Index (DSI), a lower mean value is preferable. Our findings showed a mean DSI of 0.14, suggesting enhanced drought tolerance. This is in contrast to the findings of Negisho et al. [5], who reported a higher average DSI, indicating potential advancements in drought resistance in our studied genotypes. This discovery opens avenues for the development of new wheat varieties with improved drought tolerance. Additionally, our Tolerance Index (TOL) recorded a mean of 1.63, which is on the higher end, typically indicating a lower drought tolerance compared to lower values. This result advises caution in relying solely on the TOL index for assessing drought tolerance. Our study aligns with and builds upon the groundwork laid by research such as that of Bennani et al. [44] and Eid and Sabry [45], were explored drought indices in wheat extensively. The indices we identified in our study paint a reassuring picture of the drought tolerance in the evaluated genotypes. However, it is imperative to conduct further investigations and validations across various environmental conditions and genetic varieties to make definitive conclusions about their drought tolerance capabilities.

4.2. Genome-wide association study

GWAS have proven crucial in discovering MTAs and QTLs connected to several drought-related traits in the effort to uncover the genetic basis of drought tolerance in bread wheat. Our research marks a significant advancement in this area, identifying 39 MTAs ($p < 0.001$) and 23 QTLs associated with different traits, including grain yield under varying irrigation regimes and diverse drought indices. A notable finding from our study is the dominant presence of MTAs within the B genome, where 23 MTAs were identified, surpassing the A genome, which harboured 12 MTAs. This is a notable deviation from previous findings, where the A genome was often reported to contain a higher number of MTAs, followed by the B genome, as highlighted in studies by Tadesse et al. [10] and El Gataa et al. [3]. The prominence of the B genome in our results suggests a potentially underexplored source of genetic material that could be crucial for breeding drought-resistant wheat varieties. Interestingly, the D genome maintained its historically minor role, with only 2 MTAs identified, consistent with patterns observed in earlier studies. This reinforces the understanding that the D genome plays a less significant role in drought tolerance compared to the A and B genomes. However, it's important to consider that the D genome, despite its smaller contribution, should not be overlooked in future research. It may possess unique attributes that are beneficial for improving drought tolerance, as suggested by Elhadi et al. [46] and Devate et al. [47]. Our study contributes substantially to the existing literature on the genetic basis of drought tolerance in bread wheat, highlighting the importance of the B genome and reinforcing the need for further exploration of all genomes, including the less studied D genome, for comprehensive understanding and enhancement of drought tolerance traits. Fig. 6 shows an uneven distribution of markers across the wheat genome, which may affect the accuracy of MTA detection in regions with low marker density. Although robust statistical models were used to mitigate this limitation, we acknowledge that some MTAs may have been missed in these sparsely covered regions. Future studies will consider increasing marker coverage through more comprehensive genotyping or targeted resequencing to improve the resolution of MTA identification across the entire genome.

Our study draws significant parallels and distinctions from earlier works on the genomic locations implicated in drought resilience, underscoring the conserved as well as distinct genetic regions governing this critical trait in bread wheat. A pivotal discovery was the identification of the highest marker (*wsnp_Ex_c12127_19394952*) associated with grain yield under rain-fed conditions being located on chromosome 5B. This finding resonates well with the documentation of Kumar et al. [48], who identified the QTL "MQTL5B.5" correlating with bread-making quality traits on the same chromosome. This reciprocal finding accentuates the importance of chromosome 2D as a focal point in harbouring pivotal genes regulating yield under drought conditions, suggesting the avenue for further explorative studies to unravel potential genetic markers in this chromosomal region that could be harnessed in breeding programs. The previous study did not identify the same MTAs using the same germplasm, which can be attributed to varying weather conditions [3]. Further, our scrutiny of the Drought Susceptibility Index (DSI) revealed 8 noteworthy MTAs dispersed across a diverse chromosomal landscape encompassing 1B, 5A, 5B, and 6B. This finding stands in harmony with observations made by Ballesta et al. [49], further reinforcing the critical role these chromosomal domains play in governing drought susceptibility, and essentially echoing the consensus in the scientific community on the relevance of these regions. This alignment in findings bespeaks the conserved genetic architecture underlying the drought susceptibility index, underscoring the need for a targeted investigation into these regions for developing drought-resilient bread wheat varieties. In the landscape of Stress Tolerance Index (STI), we identified 6 significant MTAs, predominantly located on chromosomes 2A, 1B, 3B, 7B, 7A and 2D. Here, we notice a divergence from the findings of Zhao et al. [50], who associated the STI index with markers on an expanded set of chromosomes, including 1B, beside the one we identified. This discrepancy invites a deeper investigation to reconcile the differences and uncover potentially overlooked regions of interest. It may also hint at the complex and multifaceted genetic basis of stress tolerance, where different studies spotlight different sets of chromosomes, possibly owing to variations in the environmental conditions and genetic materials investigated. Furthermore, the sheer number of MTAs and QTLs identified speaks to the complex genetic architecture underlying drought tolerance in bread wheat, illustrating a rich tapestry of genetic elements that are interwoven to dictate the plant's response to drought conditions. As we forge ahead, it will be imperative to not only identify but also functionally characterize these MTAs and QTLs to delineate their precise role and exploit them in breeding programs aimed at enhancing drought tolerance in bread wheat [9,51,52]. Leveraging these findings could potentially steer us closer to realizing wheat varieties that can withstand the adversities of drought while maintaining satisfactory yield levels, a step pivotal in ensuring food security in the face of changing climatic conditions. The 23 QTLs identified in this study were based on significant MTAs detected through GWAS ($p < 0.001$). Some of these QTLs appear to be novel and have not been previously reported, suggesting new insights into the genetic basis of drought tolerance and grain yield in bread wheat under the specific conditions of this study. The QTLs identified in our study align with several previously reported genomic regions associated with drought tolerance in wheat. For example, our findings are consistent with the QTLs reported by Shariatipur et al. [11,12] for agronomic traits, as well as those related to stress resilience reported by Tahmasebi et al. [53] and Sobhanian et al. [15]. These studies emphasize the importance of genomic regions related to drought response mechanisms, such as those involved in root architecture, osmotic regulation, and stomatal conductance, which may also be reflected in the QTLs identified in our study.

In our study, several SNPs were significantly associated with grain yield under both irrigated and rain-fed conditions. These findings align with previous studies, such as Shariatipur et al. [11] and Salarpour et al. [16], who identified SNPs linked to grain yield in wheat under drought and heat stress conditions. For instance, the SNP "wsnp_Ex_c12127_19394952" identified on chromosome 5B in our analysis has been associated with grain yield in similar environments, further supporting its role in drought tolerance and yield improvement. Similarly, markers identified on chromosomes 2A and 7B in our study

correspond to regions previously reported by Tahmasebi et al. [53] and Sobhanian et al. [15], who also found associations between these regions and key agronomic traits. Additionally, markers such as Kukri_c51101_351 on chromosome 7B were identified in relation to STI and grain yield under rain-fed conditions, highlighting its potential role in drought tolerance across different environmental conditions. These findings are consistent with previous studies, such as those by Shariatipur et al. [11] and Tahmasebi et al. [53], who also reported SNPs linked to grain yield and drought indices on these chromosomes. The consistency of these SNPs across multiple studies underscores their importance in marker-assisted breeding programs aimed at improving grain yield under drought stress. Furthermore, the identification of novel SNPs in our study, particularly in regions not previously reported, highlights new potential targets for enhancing drought tolerance and yield stability in bread wheat.

Epistatic interactions between markers play a critical role in determining the expression of complex traits, such as grain yield and drought tolerance. In our study, we identified several significant epistatic interactions between markers that were associated with yield-related traits. These interactions suggest that the genetic control of yield is not solely dependent on individual markers but also on the interactions between them. For example, the interaction between markers "BobWhite_rep_c66990_294" and "RAC875_c583_391" exhibited a strong multiplicative interaction effect, which was closely linked to improved yield performance under drought conditions. This suggests that these interacting loci may act synergistically to enhance the plant's resilience to water scarcity. Similarly, we observed interactions between markers on chromosome 5B and 7A that were significantly associated with both drought susceptibility and yield stability indices (YSI). These interactions likely contribute to the plant's ability to maintain stable yields under fluctuating environmental conditions, further supporting the importance of epistatic regulation in complex traits like drought tolerance. The identification of these epistatic interactions provides valuable insights into the multifactorial nature of yield determination in wheat. By uncovering how these genetic interactions influence traits, we can better understand the underlying mechanisms that allow certain genotypes to perform well under stress. Future breeding programs may benefit from targeting such interactions to develop varieties that exhibit enhanced yield stability in water-limited environments [54,55].

Several candidate genes identified in our study have been reported in previous research, reinforcing their relevance in drought tolerance and yield improvement. For instance, the TraesCS5B02G500900 gene, associated with drought tolerance in our study, was also identified by Shariatipur et al. [11] as being linked to drought-related traits in wheat. Similarly, the gene TraesCS2A02G066100**,** associated with grain yield in our study, has been highlighted in previous studies, such as Sobhanian et al. [15], for its role in yield stability under stress conditions. These overlaps suggest that these candidate genes play a crucial role in both drought response and yield maintenance, making them reliable targets for breeders aiming to improve wheat performance in water-scarce environments. By validating these genes across multiple studies, breeders can prioritize these genomic regions for marker-assisted selection and breeding programs.

An important factor influencing the accuracy of genomic prediction is the size of the training population used to calibrate the prediction models. In our study, a relatively small population was used, with 50 genotypes as the breeding population and 148 genotypes as the training population. While the results obtained offer valuable insights, the smaller population size likely imposed some limitations on the genomic prediction accuracy. Previous studies have shown that larger populations tend to enhance the precision of genomic predictions, as they capture more genetic diversity and provide more reliable estimates of marker effects. Given the relatively small size of our training population, the genomic prediction accuracy values observed (ranging from 0.31 to 0.50) should be interpreted with caution. These values may have been higher with a larger training population. Nonetheless, the observed prediction accuracies, particularly for traits such as TOL (0.50) and GY under irrigated conditions (0.43), indicate that even with a smaller dataset, meaningful and useful genomic predictions were obtained. Future studies could benefit from expanding the training population size to improve prediction accuracy and further validate the genetic markers identified.

5. Conclusion

This investigation has brought to light several genotypes exhibiting promising performance in terms of drought indices and substantial grain yields. These elite genotypes have rightfully earned a place in the global nurseries facilitated by ICARDA, positioning them as pivotal assets for our national partners in the CWANA and SSA regions. They are primed for both releases to foster enhanced agricultural outcomes and potential incorporation as foundational elements in the breeding programs, thereby steering forward the vital mission of augmenting drought resilience in bread wheat. Within the scope of our study, utilizing a robust pool of 198 spring bread wheat genotypes enabled us to pinpoint 39 MTAs and isolate 14 genes intimately associated with grain yield in both irrigated and rain-fed environments, as well as in the diverse panorama of drought indices. This rich harvest of genetic insights lays a substantial groundwork for the next strides in bread wheat research and development.

CRediT authorship contribution statement

Zakaria El Gataa: Conceptualization, Formal analysis, Methodology, Software, Writing – original draft. **Alemu Admas:** Validation, Visualization, Writing – review & editing. **Samira El Hanafi:** Validation, Visualization, Writing – review & editing. **Zakaria Kehel:** Validation, Visualization, Writing – review & editing. **Fatima Ezzahra Rachdad:** Validation, Visualization. **Wuletaw Tadesse:** Methodology, Resources, Supervision, Validation, Visualization, Writing – review & editing.

Conflicts of interest

The authors declare that there are no conflicts of interest.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.cropd.2024.100084.

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