

## RESEARCH ARTICLE

# Reaction of the Tunisian improved durum wheat cultivars to main fungal foliar diseases under field conditions

Sahbi Ferjaoui<sup>1\*</sup>, Sameh Boukef<sup>2</sup>, Cyrine Robbana<sup>3</sup>, Sebei Abdennour<sup>4</sup>, and Chahine Karmous<sup>5</sup><sup>1</sup>University of Jendouba, Regional Field Crops Research Center of Beja (CRRGCB), 9000 Beja, Tunisia.<sup>2</sup>University of Sousse, High Institute of Agronomy of Chott Mariam, 4042 Sousse, Tunisia.<sup>3</sup>National Gene Bank (BNG), 1080 Tunis, Tunisia.<sup>4</sup>University of Carthage, National Institute of Agronomic Research of Tunisia (INRAT), 2080 Ariana, Tunisia.<sup>5</sup>University of Carthage, National Agronomic Institute of Tunisia (INAT), 1082 Tunis, Tunisia.

\*Corresponding author (ferjaouisahbi@yahoo.fr)

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## ABSTRACT

Septoria tritici blotch (STB), tan spot (TS), stripe rust or yellow rust (YR), and brown rust or leaf rust (LR) are the major fungal leaf diseases reducing the productivity and quality of durum wheat (*Triticum turgidum* L. subsp. *durum* (Desf.) van Slageren) in Tunisia. Hence, screening of host plant resistance against these pathogens was the main objective of this study. A total of nine commercial Tunisian durum wheat cultivars were evaluated to STB, TS, YR and LR diseases at adult plant stage over four cropping seasons. The cvs. Karim, Razzak, Om Rabiaa, Khiar, and Maali were vulnerable to STB disease and classified as susceptible to highly susceptible (DS > 60%). While the recent cvs. Nasr, Salim, Monastir and INRAT100 were considered moderately resistant (20% < DS < 40%). Against TS, all cultivars were found to be moderately susceptible to highly susceptible (DS > 40%), except 'Khlar' which exhibit moderately resistant reaction. To YR disease, 'Khlar', 'Nasr', 'Salim', and 'Monastir' were moderately resistant, 'Karim', 'Razzak', 'Om Rabiaa', and 'INRAT100' were moderately susceptible (40% < DS < 60%), while 'Maali' had moderately susceptible to susceptible reaction (40% < DS < 80%). For LR, the cultivars were found within the range of moderately resistant to moderately susceptible (20% < DS < 60%), except 'Nasr' was resistant to moderately resistant (DS < 40%). None of the cultivars were resistant to one or more diseases. 'Karim' and 'Maali' were found as the more susceptible and 'Nasr' as the less susceptible to the four diseases. Results from this study provide valuable information that helps farmers in selecting the right wheat cultivars from the existing germplasm and for wheat breeders to enhance disease resistance in the future crosses.

**Key words:** Durum wheat, leaf rust, septoria tritici, severity, tan spot, *Triticum turgidum* subsp. *durum*, yellow rust.

## INTRODUCTION

Durum wheat (*Triticum turgidum* L. subsp. *durum* (Desf.) van Slageren) is an important crop for food security in North African region, and it is deeply rooted in the region's history and traditions. The milling of durum wheat kernel produces semolina, the raw material for pasta and the fabrication of local dishes and products including couscous, as well as various kinds of unleavened breads, bulgur and frike. In Tunisia the cultivated area of durum wheat has reached about 0.54 million ha with production around 0.95 million tons (ONAGRI, 2021). The gap between increasing demand and current production is a big challenge for wheat breeders and scientists. This challenge can be met by increasing the cultivated area and yield per unit area and minimizing the production losses. Expanding area seems almost impossible because of limited available area and other constraints like drought, urbanization, and trend in fruit trees.

In the last two decades, there have been increasing problems with climatic changes and onset of severe plant fungal diseases epidemics of cereals worldwide. Indeed, the wheat crop is attacked by many diseases throughout the growing season, of which leaf spot and rusts are of great importance in most wheat growing

regions worldwide. This has been attributed to changes in pathogen populations, evolution and spread of new virulent and adapted races to diverse agricultural conditions (Singh et al., 2016). The factors favoring the spread of cereal pathogens are the traditional seed exchanges, as well as climate change, and the transmission of new variants of pathogens by air which contributes to an increase in the geographical range of pathogenic species (Udayanga et al., 2020). While the occurrence of fungal diseases is influenced by agronomic management practices such as reduced tillage, limited crop rotation, monoculture, high N fertilization, crop residues left in the field and susceptible cultivars.

In North African countries, especially in Tunisia, four major fungal leaf diseases can be found in durum wheat, including septoria tritici blotch (STB) caused by the fungus *Zymoseptoria tritici* (formerly known as *Mycosphaerella graminicola*), tan spot (TS) caused by *Pyrenophora tritici-repentis* (Died.), leaf rust (LR) or brown rust caused by *Puccinia triticina* f. sp. *tritici* Erikss (*Pt*) and stripe rust or yellow rust (YR) caused by *Puccinia striiformis* f. sp. *tritici* Erikss (*Pst*). The occurrence and severity of these diseases have increased in the last decade, partly due to climate change (Singh et al., 2023). On average, yield losses due to these pathogens may reach up to 50% during epidemic seasons by reducing grain size, grain weight, number of grains per head, total biomass, and grain quality (Ferjaoui et al., 2023).

The most common method of controlling these pathogens is the application of fungicides. However, selection for fungicide resistance or tolerance due to repeated and irrational use has been reported (Fisher et al., 2018). Therefore, combining correct cultural practices including proper crop rotation with cultivars with good resistance levels, can reduce the occurrence of fungal diseases and contributes to save fungicide treatments. For all wheat diseases, genetic resistance emerges as an indispensable component in disease management, because it is cost-effective, environmentally sustainable, and valuable to farmers who have no access to fungicides.

In Tunisia, durum wheat breeding program was started in the early 20th century, from French colonists who rounded up the local landraces (Martínez-Moreno et al., 2020), particularly the lines Derbassi, Biskri, Adjimi, and Jenah Khottifa which were dominant. From these populations, the landrace Mahmoudi was selected based on their production potential and became the dominant durum wheat variety until the 1960s. From 1940 to 1970 more productive and better disease-resistant old varieties were selected from introduced seeds or local crosses and registered in the official Tunisian catalogue of varieties, such as Chili-931 in 1953, Kyperounda in 1954, INRAT-69 and Badri in 1969. By the early 1970s, semidwarf durum lines were developed by International Maize and Wheat Improvement Center (CIMMYT) and International Center for Agricultural Research in the Dry Areas (ICARDA) allowed the selection and registration of new yielding cultivars, more adapted to rainfed conditions, among of them Karim, Khiar, Razzak, and Om Rabiaa. Starting from the 20th century, breeding efforts had been undertaken by the national durum wheat breeding program, in collaboration with CIMMYT and ICARDA to further improve wheat characteristics particularly resistance to fungal diseases. Consequently, new high-yielding modern durum wheat varieties more suitable and adapted to intensive production systems were released (Gharbi and El Felah, 2013; Martínez-Moreno et al., 2020).

Despite the ongoing efforts made by breeders to develop and promote disease resistant new durum wheat cultivars, Tunisian farmers complain about the susceptibility of the used wheat varieties to leaf fungal diseases. Therefore, the objective of this study was to determine the reaction of nine Tunisian cultivated durum wheat cultivars to STB, TS, YR and LR, and to estimate the disease severity (DS) on different wheat cultivars under field conditions.

## MATERIALS AND METHODS

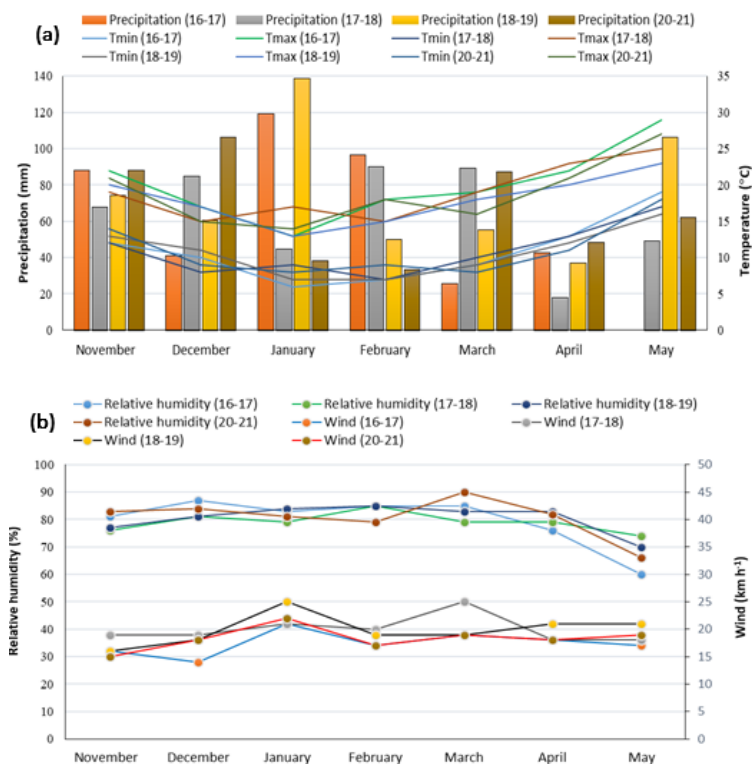
### Plant material and trial management

Nine improved durum wheat (*Triticum turgidum* L. subsp. *durum* (Desf.) van Slageren) varieties including eight varieties obtained from national wheat breeding program and one released variety introduced by the private sector were used in the present study (Table 1). The field experiments were performed in the northwestern Tunisia, at the experimental station of the Regional Field Crops Research Center of Beja (CRRGCB) during four wheat growing seasons (2016-2019 and 2020-2021). This region is part of sub-humid bioclimatic zone, characterized by an average annual precipitation that ranges between 500 and 850 mm and a daily mean temperature ranging between 10 and 28 °C. Average monthly precipitation, minimum and maximum air temperatures, as well as relative humidity and wind speed were collected for all years of the study (Figure 1)

from weather station located at a direct distance of 300 m from the research plots. The experiments were set up as a randomized complete block design (RCBD) with four replicates per treatment, spaced 1.5 m apart. Plots were three rows 1.5 m long with 20 cm spacing, with sowing density of 60 seeds per row. The spacing between plots was 0.5 m. Trials were established at the regular sowing date in the beginning of December 2016, 2017, 2018 and 2020. To ensure adequate crop development, fertilization (N, P<sub>2</sub>O<sub>5</sub>, and K<sub>2</sub>O) and weed control were conducted according to standard experimental practices in the station.

**Table 1.** Origin, year of release and pedigree of durum wheat varieties. INRAT: National Institute of Agronomic Research of Tunisia; CIMMYT: International Maize and Wheat Improvement Center; ICARDA: International Center for Agricultural Research in the Dry Area.

Variety name	Origin	Year of release	Cross name, pedigree
Karim	Introduced from CIMMYT in 1973 (F4), selected by INRAT	1980	D21563/AA"S"/FG"S"
Razzak	Cross by INRAT in 1976, CIMMYT parent	1987	21563/AO//FG/3/DM//69/331
Khlar	Cross by INRAT	1992	Chen"S"/Altar 84
Om Rabiaa	INRAT and ICARDA	1996	Jori C69/Hau
Nasr	INRAT and ICARDA	2003	GdoVZ512/Cit//Ruff/Fg/3/Pin/Gre//Trob
Maali	INRAT	2007	CMH80A.1016/4/TTURA/CMH74A370/CMH77.774/3/YAV7 9/5/Razzak/6/DACK"S"/YEL"S"/Khlair
Salim	INRAT	2009	Altar84/FD84-19-126-1-2/Razzak/3/Krf/Baladia Hamra
Monastir	RAGT Semences (France)	2012	-
INRAT100	INRAT and CIMMYT	2016	CDSS07Y00779D-2B-02Y-07M-5Y-0B-8Bj-3Bj-0Bj



**Figure 1.** Precipitation, minimum and maximum air temperatures (a), relative humidity and wind speed (b) recorded during four cropping seasons (2016-2017, 2017-2018, 2018-2019 and 2020-2021) from weather station, close to the experimental site.

### Fungal material and inoculation

The nine cultivars were subjected to field evaluation for Septoria tritici blotch (STB), tan spot (TS) and stripe rust or yellow rust (YR) during 2016-2017, 2017-2018 and 2018-2019 growing seasons under natural conditions as well as using artificial inoculation, whereas in 2020-2021 evaluation was carried out under natural infection. For resistance to brown rust or leaf rust (LR), evaluation in all trials was based on natural infection. To initiate primary infection of wheat cultivars by STB and TS, experimental plots were covered with infested wheat straw and stubble debris from the previous cropping. For STB resistance, the artificial inoculum was a mixture of three virulent and aggressive isolates of *Zymoseptoria tritici* (TunBz-1, TunBj-13, TunM-1) that were previously characterized by Ferjaoui et al. (2015). The spore's suspension was produced on yeast-glucose liquid medium (yeast extract 10 g L<sup>-1</sup>, glucose 30 g L<sup>-1</sup>), as described by Ferjaoui et al. (2015), and adjusted to a concentration of 10<sup>7</sup> spore mL<sup>-1</sup> to which drops of the detergent Tween 20 (polyoxyethylene sorbitan monolaurate) were added (1 drop 100 mL<sup>-1</sup>) prior to inoculation. Trials were inoculated twice at tillering stage (Zadocks growth stage (ZGS) 21) (Zadocks et al., 1974) and at stem elongation (ZGS 31) using a backpack-carried manual sprayer.

For evaluation to TS resistance, the *Pyrenophora tritici-repentis* isolate representing the frequent and prevalent race 7 in the main wheat growing-area of Tunisia (Kamel et al., 2019; Laribi et al., 2019) was used for inoculation. This isolate was grown on V8-potato dextrose agar (V8-PDA) following the protocol described by Kamel et al. (2019). The inoculum concentration was adjusted to 3000 conidia mL<sup>-1</sup> and supplemented with one drop of Tween 20 surfactant 100 mL<sup>-1</sup> conidial suspension before inoculation, used to avoid the aggregation and helps an equal distribution of the suspension over the plants. Wheat plots were inoculated at tillering stage (ZGS 25), by spraying conidia suspension until run-off, and repeated after 10 d.

For reaction to YR, in addition to natural infection, the 'Warrior 3' race was used for artificial inoculation. This strain was collected in Tunisia in 2014 from infected durum wheat grown in the same experiment station, characterized and multiplied by the research unit BIOGER-CPP (Biologie, Gestion des Risques en Agriculture - Champignons Pathogènes des Plantes) of National Institute for Agricultural Research (INRA), France (Bahri et al., 2016). The inoculum was provided in the form of uredospores stored in Eppendorf tubes by the Laboratory of Bioaggressors and Integrated Protection in Agriculture at the National Agronomic Institute of Tunisia. One row of wheat highly susceptible line 'Victo' was sown between blocks to act as YR spreader and ensure optimal disease distribution among and within plots. The inoculum was applied at stem elongation stage (ZGS 30), directly to the upper leaves of the spreader line using a cotton swab. To ensure a good infection, a second inoculation was repeated after 2 wk according to the same method.

### Disease assessment

The response of wheat cultivars to STB and TS was evaluated at flowering stage (ZGS 61). Disease severity (DS) was estimated visually on whole plants of each plot using the modified double-digit (00-99) scale (Eyal et al., 1987). The first digit (D<sub>1</sub>) indicates disease progress in canopy height from the ground level (0-9), while the second digit (D<sub>2</sub>) refers to severity measured as the diseased leaf area, using a scale from 0 to 9. The DS percentage was estimated using the following formula (Duveiller et al., 2005):

$$DS \% = (D_1/9) \times (D_2/9) \times 100$$

For LR and YR diseases, severity was assessed when susceptible plots displayed maximum level of plant infestation, corresponding to late milk development stage (ZGS 71) for YR and early dough development stage (ZGS 83) for LR. The severity estimation was based on 0% to 100% modified Cobb's scale (Peterson et al., 1948).

Based on DS values calculated from corresponding double-digit scores, classification of wheat cultivars was made following severity classes: Resistant (R) for DS from 0% to 20%, moderately resistant (MR) 21% to 40%, moderately susceptible (MS) 41% to 60%, susceptible (S) 61% to 80%, and highly susceptible (HS) DS > 80%.

### Data analysis

The ANOVA of collected data was effectuated using the general linear model (GLM) procedure of SAS software (SAS Institute, Cary, North Carolina, USA). Agglomerative hierarchical clustering (AHC), using the unweighted pair group method with the arithmetic mean method (UPGMA), was performed to rank the genotypes according to their DS scores. The clusters were subsequently generated using R statistical software version 4.2.1. (R Foundation for Statistical Computing, Vienna, Austria).

## RESULTS

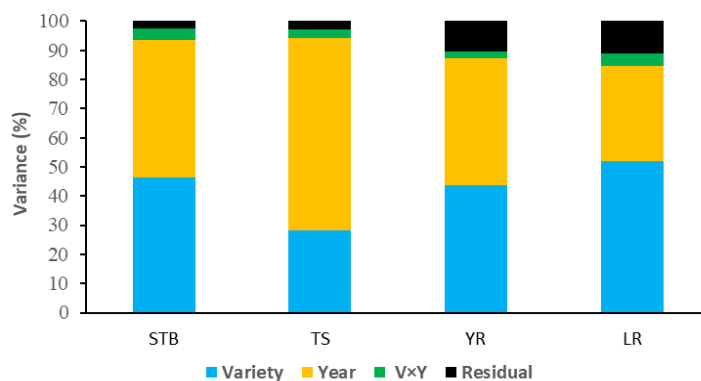
### Screening for resistance to leaf spot and rust diseases

The ANOVA for disease severity (DS) showed that DS was significantly ( $P < 0.001$ ) affected by variety (V) and year (Y). The  $V \times Y$  interaction was significant ( $P < 0.001$ ) for STB, TS, and LR ( $P < 0.05$ ) severities, except for YR (Table 2).

Results showed that V and Y were the major contributor sources of DS variability for STB (46.48% and 46.86%, respectively) and YR (43.82% and 43.46%, respectively). However, TS severity was mainly related to Y variance (65.93%). In contrast, LR severity was dependent on V (52.10%). The  $V \times Y$  contribution to all disease severities was lower and almost the same for all evaluated diseases (Figure 2).

**Table 2.** ANOVA with mean squares for disease severity on nine improved durum wheat varieties evaluated during four cropping seasons. STB: Septoria tritici blotch; TS: tan spot; YR: yellow rust; LR: leaf rust. \* $P < 0.05$ ; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$ ; NS: nonsignificant.

Source of variation	STB severity	TS severity	YR severity	LR severity
Variety (V)	4510.924***	2368.616***	1554.861***	1510.938***
Year (Y)	12127.557***	14781.756***	4111.806***	2524.074***
$V \times Y$	137.521***	84.732***	28.472 <sup>NS</sup>	39.178*
R <sup>2</sup>	0.968	0.962	0.864	0.852

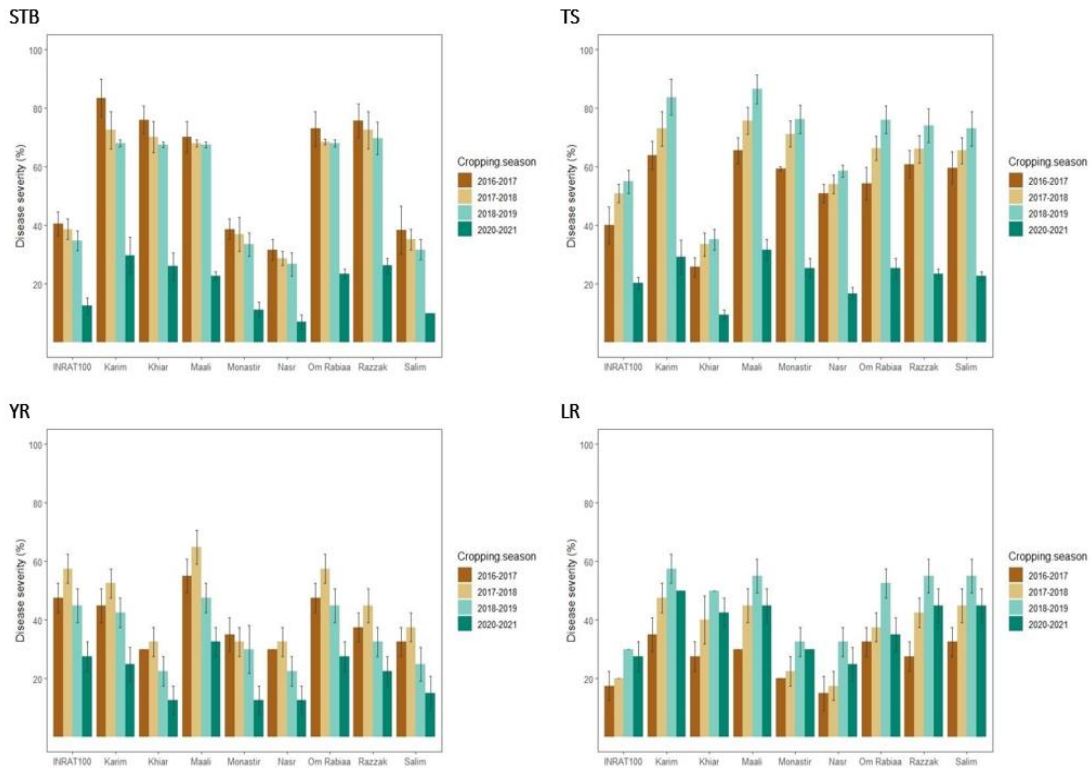


**Figure 2.** Partitioning of total variance for septoria tritici blotch (STB), tan spot (TS), yellow rust (YR), and leaf rust (LR) severities into variety (V), years (Y),  $V \times Y$  interactions, and residual variance components.

### Evaluation against septoria tritici blotch and tan spot

The DS averages of the nine wheat cultivars are presented in Figure 3. From 2016-2017 to 2018-2019 cropping seasons, there was heavy STB and TS disease pressure as indicated by a wide range, and high levels of DS (from 25.75% to 86.50%). Climatic conditions were relatively more conducive to disease development over these seasons. Indeed, important rainfall and relative humidity levels were recorded in December, January, and February (Figure 1) supporting plant infection and the dissemination of the secondary inoculums.

Durum wheat cultivars were classified for their response to different diseases according to their percentage of DS and using the scale aforementioned. For STB disease, Karim was the most susceptible cultivar, with an average DS ranging from 68.00% to 83.50% (Figure 3). Similarly, 'Razzak', 'Khlar', 'Om Rabiaa', and 'Maali' showed S reaction with a DS higher than 60.00% and less than 80.00%. However, 'INRAT100' showed MR to low susceptibility across cropping seasons to STB with a DS reaching a maximum of 40.50% in 2016-2017 (Figure 3). 'Monastir', 'Nasr', and 'Salim' were classified as MR to STB with a DS ranging from 20.00% to less than 40.00% (Table 3; Figure3).



**Figure 3.** Disease severity pattern of septoria tritici blotch (STB), tan spot (TS), yellow rust (YR), and leaf rust (LR) on nine improved durum wheat varieties under field conditions, evaluated during four cropping seasons.

**Table 3.** Mean disease severity (DS) range of septoria tritici blotch (STB), tan spot (TS), yellow rust (YR), and leaf rust (LR) on nine durum wheat cultivars and their disease reaction recorded during favorable cropping seasons (from 2016-2017 to 2018-2019 for STB, TS and Yr; 2017-2018, 2018-2019 and 2020-2021 for LR). R: Resistant; MR: moderately resistant; MS: moderately susceptible, S: susceptible; HS: highly susceptible.

Cultivar	STB		TS		YR		LR	
	DS range	Disease reaction	DS range	Disease reaction	DS range	Disease reaction	DS range	Disease reaction
	%		%		%		%	
Karim	68.00-83.50	S-HS	63.80-83.70	S-HS	42.00-52.00	MS	47.00-57.00	MS
Razzak	69.00-75.75	S	60.90-74.00	S	32.50-45.00	MR-MS	42.00-55.00	MS
Khair	67.50-76.00	S	25.70-35.20	MR	22.50-32.50	MR	40.00-50.00	MS
Om Rabiaa	68.00-73.00	S	54.20-76.00	MS-S	45.00-57.00	MS	35.00-52.50	MR-MS
Nasr	26.70-31.50	MR	50.90-58.50	MS	22.50-32.50	MR	17.50-32.00	R-MR
Maali	67.50-68.00	S	65.50-86.50	S-HS	47.50-65.00	MS-S	45.00-55.00	MS
Salim	31.70-38.30	MR	59.60-73.00	MS-S	25.00-37.50	MR	45.00-55.00	MS
Monastir	33.50-38.70	MR	59.30-76.20	MS-S	30.00-35.00	MR	22.50-32.50	MR
INRAT100	34.70-40.50	MR-MS	40.10-54.90	MS	45.00-57.00	MS	20.00-30.00	MR

Results of TS showed that 'Karim' and 'Maali' were HS with a DS exceeding 80.00%. They were followed by 'Razzak' with DS less than 80.00%. However, the reaction type of 'Om Rabiaa', 'Salim', and 'Monastir' stretched from MS to S with DS values varying from more than 50.00% to less than 80.00%. 'Nasr' and 'INRAT100' exhibited a DS ranging from 40.00% to 60.00% and were classified as MS. Besides, 'Khiair' was found to be MR to TS with a DS lesser than 40.00% (Table 3, Figure 3).

It is important to notice that during the cropping season 2020-2021, a lower infection level was observed for both STB and TS with a DS lesser than 30.00% even on the susceptible cultivars. The observed results could be attributed to the absence of artificial inoculation, and unfavorable environmental conditions as deficient rainfall recorded during January and February (Figure 1), which reduced natural infection, development, and spread of the disease.

#### **Evaluation against yellow rust and leaf rust**

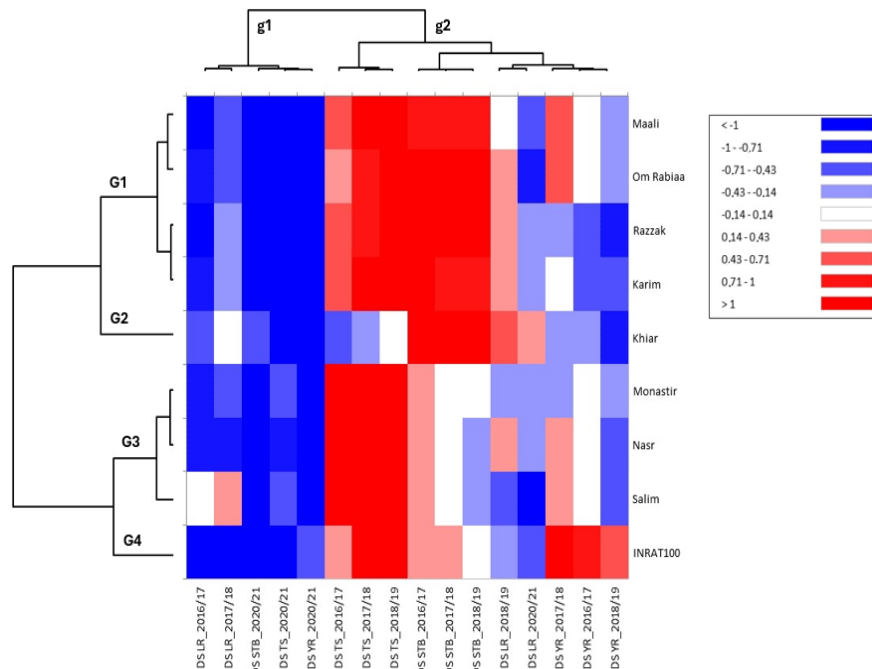
The evaluation of durum wheat reaction to YR showed DS levels lower than those recorded for STB and TS diseases. Despite the favorable environmental conditions for the development of YR disease from 2016-2017 to 2018-2019 seasons, only 'Maali' showed a reaction ranging between MS and S ( $40.00\% < DS < 80.00\%$ ). However, 'Om Rabiaa', 'INRAT100', and 'Karim' were classified as MS to YR with DS ranging from more than 40.00% to less than 60.00%. While 'Razzak' with DS varying from 32.50% to 45.00% was classified from MR to MS. Besides, 'Khiair', 'Nasr', 'Monastir', and 'Salim' were found to be MR with a DS ranging from 22.50% to 37.50% (Table 3, Figure 3). During the 2020-2021 cropping season, low rainfall and relative humidity during January and February combined with high temperatures in Mars and April (Figure 1), affected YR development and expression of symptoms. Consequently, DS decreased and did not exceed 27.50% on the susceptible genotypes (Figure 3).

The LR occurrence was more important under weather conditions of 2018-2019 and 2020-2021 cropping seasons and with a lesser degree in 2017-2018. Indeed, considerable amounts of rainfall, high humidity, and ideal air temperature were registered in Mars, April and May 2019 and 2021 (Figure 1), enhancing natural infection and expression of disease symptoms. During the same period, 'Karim', 'Razzak', 'Khiair', 'Maali', and 'Salim' showed MS reaction and DS average ranging from 40.00% to less than 60.00%. Whereas 'Om Rabiaa' exhibited MR to MS reactions with a DS ranging between 35.00% and 52.50%. 'Monastir', and 'INRAT100' revealed MR reaction with DS values less than 33.00% (Table 3, Figure 3). Moreover, 'Nasr' showed a reaction ranging between R and MR. In contrast during 2016-2017 cropping season, climatic conditions characterized by relatively high temperatures, and low rainfall and humidity (Figure 1), lead to low DS ( $< 35.00\%$ ) on the susceptible cultivars.

#### **Hierarchical classification of durum wheat cultivars based on the disease reactions**

Durum wheat cultivars were clustered based on mean DS values calculated for each disease. Cluster analysis based on ascendant hierarchical clustering (AHC) led to the separation of the tested cultivars into four groups (Figure 4). The first group (G1) was constituted by 'Maali', 'Om Rabiaa', 'Razzk', and 'Karim' which positively correlated (red color) and had a similar behavior to STB and TS. These cultivars showed disease reactions ranging from HS to MS (Table 3). The second group (G2), represented by 'Khiair' with susceptibility to STB and LR, and MR reaction to TS and YR (Table 3). The third group (G3) encompassed 'Monastir', 'Nasr', and 'Salim' which showed MR reaction to STB and YR, S to MS reaction to TS, and MS to R reaction to LR (Table 3). The fourth group (G4) composed by 'INRAT100' which showed MR reaction to STB and LR, and MS reaction to TS and YR.

Likewise, the severity of different diseases across cropping seasons was clustered into two groups. The first group 1 (g1) contains the DS recorded during unfavorable cropping seasons (2016-2017 and 2017-2018 for LR, and 2020-2021 for STB, TS, and YR). In contrast, the second group (g2) encloses the DS obtained during favorable cropping seasons (2016-2017 to 2018-2019 for STB, TS, and YR, and 2018-2019 to 2020-2021 for LR).



**Figure 4.** Heatmap and hierarchical clustering of the nine improved durum wheat varieties based on the disease severity (DS) across four cropping seasons (2016-2017 to 2018-2019, and 2020-2021). Red and blue colors indicate high positively or negatively correlations for the corresponding trait, respectively. Darker the hue greater the relation between the traits and vice versa. DSLR, DSSTB, DSYR, DSTS indicate disease severity for leaf rust, septoria tritici blotch, yellow rust and tan spot, respectively.

## DISCUSSION

Fungal leaf diseases have a devastating impact on wheat production, and their frequency and severity are increasing due to global climate change (Singh et al., 2023). To overcome these challenges, the use of cultivars with high disease resistance will ensure stable yield and steady revenue for farmers. Thus, to succeed in breeding programs it is crucial to evaluate precisely the wheat disease response which is considered an important selection criterion. The present study evaluated the severity of STB, TS, Yr, and LR diseases on nine Tunisian durum wheat cultivars across four cropping seasons. Nayak et al. (2018) noted that the disease severity (DS) in plant-pathosystems can be assessed either once at the peak of the epidemic development (Kamel and Cherif, 2021; Iqbal et al., 2022) or several times with intervals from the beginning till the end of the epidemic (Ferjaoui et al., 2015). To obtain accurate assessment results in the present investigation, DS scoring started from the flowering growth stage (ZGS 61 for STB and TS, ZGS 71 for YR, and at ZGS 83 for LR) when the maximum of disease symptoms on wheat leaves was observed on the susceptible cultivars.

The results showed different responses of wheat cultivars across diseases and cropping seasons. These observations could be due to the climatic condition effects (Y), as reported by Gafencu et al. (2018). Thus, the accurate disease resistance assessment should be released across multiple cropping seasons at disease hotspots and under favorable environmental conditions. Moreover, to enhance disease infection, cultivars were evaluated under both natural infection and artificial inoculation conditions using available virulent and predominant isolates and races of the pathogens, except for leaf rust. All these factors can influence the DS (Fagundes et al., 2020). Besides, the tested wheat cultivars significantly affected the reaction to all tested pathogens. Indeed, the cultivars showed a varying reaction regarding the severity of diseases with moderately resistant to highly susceptible. The observed variation is mainly accounted to the contribution of the variable genetic background of the tested cultivars.



Results obtained from the evaluation against STB indicated that most of the cultivars were MR to S. Highest DS was recorded on 'Karim', 'Khiar', and 'Razzak', which confirmed previous finding of Bel Hadj Chedli et al. (2018). The susceptible 'Karim', 'Khiar', and 'Om Rabiaa' were used as susceptible controls in previous studies (Ferjaoui et al., 2015; 2022; Ben M'Barek et al., 2022; Ferjaoui et al., 2023). 'Maali' found as S, was characterized as resistant at the time of its release in 2007 (Gharbi and El Felah, 2013). Our results are in agreement with those of Hassine et al. (2018), who reported that 'Karim', 'Razzak', and 'Maali' exhibited the highest area under disease progress curve (AUDPC) compared to 'Salim', recorded under natural conditions during 2014-2015 season at Oued-Beja location. The reaction of 'Salim' confirms the result obtained by Berraies et al. (2011), which showed that this cultivar is a source of partial resistance to STB and harbors one quantitative trait loci (QTL) located on chromosome 3B. 'Nasr' and 'Salim' were tested with a wide collection of Mediterranean durum wheat landraces in Tunisia during the 2016-2017 cropping season under field conditions and using infected wheat-straw and artificial inocula of five mixed *Zymoseptoria tritici* strains (Ben M'Barek et al., 2022). The obtained results ( $0.4 < \text{relative AUDPC} < 0.6$ ) confirmed the MR reaction of these cultivars. In contrast, the imported released 'Monastir' and the recently improved 'INRAT100' which recorded MR, and MR to MS, respectively, were previously characterized as R (Bel Hadj Chedli et al., 2018).

The response to TS ranged from MS to S, except 'Khiar' which showed a MR reaction. Our results are in agreement with previous studies which revealed the MS of 'Salim', 'Monastir', and 'INRAT100' (Kamel and Cherif, 2021; Laribi et al., 2022a), and the susceptibility of 'Karim', 'Razzak', and 'Maali' (Kamel and Cherif, 2021; Laribi et al., 2022a; Tissaoui et al., 2022). In contrast, some differences were observed with other studies that classified 'Nasr' and 'Khiar' as S, and 'Salim' as R (Laribi et al., 2021; 2022a, Tissaoui et al., 2022).

Differences in the response of some cultivars to diseases could be attributed to (i) agronomic (cultural practices), and environmental (fluctuating climatic elements) factors which strongly correlated to the severity of the disease (Hailemariam et al., 2020), (ii) evaluation method and the adopted classification scale, and (iii) the coexistence of different forms of the pathogen involved in the infection even in the same growing season and in the same field. In this context high genetic diversity was reported in the Tunisian STB (Bel Hadj Chedli et al., 2022) and TS (Laribi et al., 2022b) populations, which may lead to different levels of disease severity on the host.

The YR disease is found almost every year in Tunisia wherever wheat is cultivated. The DS evaluation showed that the most cultivated cultivars such as Karim, Maali and Razzak were MS to S. Despite some cultivars were MR (Khiar, Nasr, Salim, and Monastir), this pathogen become a serious threat to Tunisian durum wheat germplasm because it is able to evolve new races and virulence that quickly overcome introduced resistance. Indeed, high temperature-adapted aggressive strains were detected in North Africa and in Tunisia (Warrior and triticale races) (Bahri et al., 2016). For LR, among the nine cultivars six were MS and the rest was MR. Aoun et al. (2019) reported that the large majority of durum wheat cultivars are susceptible to the durum leaf rust races. This result highlight that this pathogen is a problematic since it displays high diversity and exhibits high adaptability to a wide range of climates (McCallum et al., 2016). Therefore, there is an urgent need to identify novel resistance sources and develop new durum wheat cultivars since few cataloged Lr genes have been globally deployed in breeding programs thus far (Aoun et al., 2019). Despite the importance of YR and LR diseases in the Tunisian durum wheat fields during the last decade, no reports aimed at characterizing the cultivated cultivars against these pathogens. Therefore, our study is the first investigation highlighting the behavior of the released Tunisian durum wheat cultivars against these diseases.

Wheat breeding can produce resistance to individual diseases, but it is challenging to select genetic resistance against multiple diseases simultaneously. Partial resistance has become an important aim for wheat breeding programs because it is often durable and reduced level of epidemic development (Martínez-Moreno et al., 2022). Our data showed that 'Monastir', 'Nasr', 'Salim' (G3), 'INRAT100' (G4), and 'Khiar' (G2) had at least partial resistance to two diseases. This reflects the importance of these cultivars despite none being highly disease resistant.

## CONCLUSIONS

This study highlighted that none of the recent or old-released durum wheat cultivars was resistant to leaf diseases. However, they were highly appreciated by farmers for their good agronomic performance and yield stability. The most common sown wheat cultivars in Tunisia (Karim, Razzak, Om Rabiaa, and Maali) were

classified as moderately susceptible to susceptible to all leaf diseases. Despite that 'Karim' and 'Maali' were the most cultivated for their productivity were observed as the most susceptible to the fungal diseases. However, the most recently released cultivars seem have a partial resistance to septoria tritici blotch (Monastir, Salim, INRAT 100), yellow rust (Salim and Monastir), and leaf rust (Monastir and INRAT 100) diseases. Although being less exploited commercially for its sensitivity to lodging, 'Nasr' seems the most suitable in preventing leaf diseases. For securing wheat production against continuously and rapidly evolving pathogens, and in response to agricultural needs, breeding resistant varieties with high agronomic performance is essential. The release of new cultivars with high level of resistance to leaf diseases can avoid disease epidemics and maintain the inoculum at lower levels. This can be achieved through the identification of novel sources of resistance that could be used as a genetic source in the wheat breeding program.

#### Author contribution

Conceptualization: S.F. Methodology: S.F., S.B., A.S. Formal analysis: S.F., C.K. Investigation: S.F., A.S. Resources: S.F., A.S. Data curation: S.F., S.B., C.R. Writing-original draft: S.F. Writing-review & editing: S.F., S.B., C.R., C.K. All coauthors reviewed the final version and approved the manuscript before submission.

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