https://doi.org/10.17221/103/2022-PPS

# Progression of wheat tan spot under different bioclimatic stages and agricultural practices

*Salma Tissaoui*[1]*, *Noura Omri Benyoussef*[2], *Hatem Chaar*[3], *Marwa Hassine*[4], *Jean-Stephane Venisse*[5], *Bouzid Nasraoui*[1], *Amira Mougou Hamdane*[1]

[1]*Laboratory of Bioggressors and Integrated Pest Management in Agriculture (LR14AGR02), National Agronomic Institute of Tunisia, University of Carthage Tunis, Tunis, Republic of Tunisia*
[2]*Field Crop Laboratory, National Agronomic Research Institute of Tunisia, Ariana, Republic of Tunisia*
[3]*Crop Improvement Laboratory, National Institute of Agronomy of Tunisia, Ariana, Republic of Tunisia*
[4]*Laboratory of Genetics and Cereal Breeding (LR14AGR01), National Agronomic Institute of Tunisia, University of Carthage, Tunis, Republic of Tunisia*
[5]*Clermont Auvergne University, INRAE, PIAF, Clermont-Ferrand, France*

**Corresponding author: salmatissaoui2@gmail.com*

**Citation:** Tissaoui S., Omri Ben Youssef N., Chaar H., Hassine M., Venisse J.S., Nasraoui B., Mougou-Hamdane A. (2023): Progression of wheat tan spot under different bioclimatic stages and agricultural practices. Plant Protect. Sci., 59: 233–244.

**Abstract:** Tan spot, caused by *Pyrenophora tritici-repentis,* is a severe worldwide wheat foliar disease and has become common in Tunisia. Two hundred wheat field surveys were conducted in northern Tunisia during the 2017–2019 seasons to determine disease intensity and the correlation of the tan spot with agricultural practices and environmental factors. The disease infested 67% of the visited regions. The highest prevalence (87.5%), incidence (> 30%), and severity (> 10%) were observed in sub-humid regions, contrary to the middle semi-arid regions. Logistic regression was used to analyse the correlation of incidence and severity with various factors as independent variables. In a multiple variable model, the bioclimatic stages, variety, previous crop, humidity, and rainfall were significantly associated with tan spot incidence and severity. High incidence and severity were significantly correlated to the sub-humid regions, varieties Maâli, Razzak, Karim, and Carioca, rainfall, high level of humidity, and monoculture. The correlation between the tan spot and several environmental factors must be considered to ensure successful and sustainable disease management strategies.

**Keywords:** correlation; fields wheat; incidence; severity; sub-humid; previous crop; variety

Cereal crops are a primary source in the diet of several world populations, such as the Maghreb countries and the Middle East (Le Mouel et al. 2015), because of the culinary traditions favouring cereal-based foods (Anonymous 2012). In Tunisia, cereal crops, namely durum wheat, common wheat, oat, barley and triticale, often occupy a third of the area sown, representing about 1.5 million ha annually (El Felah & Gharbi 2014). However, the area sown has decreased over the last decade (2010–2020) from 1.343 million ha in 2012 to 1.127 million ha in 2016 (DGPA 2020). The cereal production was also char-

Supported by the Laboratory of Bioggressors and Integrated Pest Management in Agriculture (LR14AGR02), National Agronomic Institute of Tunisia.

https://doi.org/10.17221/103/2022-PPS

acterised by an inter-annual fluctuation, with the lowest crop of 1.07 million t in 2010 and a peak crop of 2.4 million t in 2019. High production was only obtained for four seasons over the last decade (DGPA 2020). Durum wheat (*Triticum durum*) is the main cereal crop in production and area sown. The wheat area is larger than any other cereal and varies from 53% to 65% of the total cereal area. However, the cultivated area has decreased during the last decade, from 636 000 ha in 2010 to 458 000 ha in 2020. The wheat production also varied from year to year, with a minimum production of 757 600 t in 2015 and a maximum of 1.267 million t in 2019 (DGPA 2020). The wheat areas are geographically located primarily in the northern regions of Tunisia, belonging to the humid, sub-humid, upper semi-arid and middle semi-arid bioclimatic stages (Anonymous 2016; DGPA 2020). Climatic factors like total rainfall and heavy dew contribute to create favourable conditions for the occurrence of fungal diseases in wheat crops (Iram & Ahmad 2005; Kremneva & Gudoshnikova 2011) during crop development and under different growing environments (Weise 1987). The fungal diseases cause major damage (Ezzahiri 2001) to wheat crops because plant pathogen can attack roots, stem, and spikes and induce several symptoms. Tan spot is a foliar disease caused by *Pyrenophora tritici-repentis* (anamorph of *Drechslera tritici-repentis*), which is observed in several countries worldwide and has a considerable economic impact (Fernandez et al. 1998; Ali & Francl 2003; Wakulinski et al. 2003; Tadesse et al. 2006; Perelló & Dal Bello 2011). The necrotrophic fungus induces up to 53% of yield losses and reduces kernel weight, number of grains by head and total biomass (Moreno et al. 2012). *P. tritici-repentis* infects wheat plants at any growth stage and produces lesions on young wheat seedlings. Tan spot disease involves two distinct symptoms: tan necrosis and extensive chlorosis (De Wolf et al. 1998; Singh et al. 2010; Benslimane et al. 2011). Characteristic symptoms are elliptical to elongate spots, dark brown near the centre and surrounded by a yellow border. The lower, more mature leaves are infected first, and the pathogen spreads to the upper leaves as the disease progresses (Wegulo et al. 2012). The susceptible cultivars, agricultural practices as zero-tillage, shorter rotation and continuous wheat cultivation promote a high disease incidence (Ciuffetti & Tuori 1999). These factors will allow the build-up of inoculum on the wheat stubble over time (Moreno et al. 2008). Tan spot is in increase in recent years in the Mediterranean region (Andonova & Todorova 2007; Benslimane et al. 2017) and causes high damage like in Morocco where yield losses reach 18% (Gamba et al. 2017). In Tunisia, the first observation of disease was in the late 1990's when its influence was negligible on wheat crops (Cherif et al. 1994). It is an emergent disease whose occurrence and spread have progressed throughout the last decades in the Tunisian wheat fields. The disease's prevalence, incidence and severity increased in several cereals during the last five years under distinct weather conditions (Kamel 2021; Tissaoui et al. 2022). Understanding the relationship of disease intensity with environmental conditions and agricultural practices will allow determining the most significant variables useful for developing effective and sustainable phytosanitary management program (Edema et al. 1997). Therefore, this work aims (*i*) to determine incidence, and severity of wheat tan spot in several regions belonging to distinct bioclimatic stages of Northern Tunisia, and (*ii*) to evaluate the effects of bioclimatic conditions and agricultural practices on disease progression.

## MATERIAL AND METHODS

**Sampling, evaluation and identification of the disease.** Several surveys have been planned between mid-February to May, from 2017 to 2019, in different durum wheat and bread wheat fields to detect tan spot disease. The surveys were conducted when wheat crop was between the first node detectable of stem elongation (Z31) and soft dough growth stages (Z85) following the procedure described by Zadoks et al. (1974) and Stubbs et al. (1986). The visited regions belong to nine governorates of northern Tunisia as follows: Jendouba (Ghardimaou, Bousalem, Oued Mliz), Beja (Oued Beja, Guebollat, Mjez Elbab, Elaferigue, Teborsouk), Kef (Nebber, kef N/S, Eddir, Boulifa), Zaghouan (Zaghouan N/S, Bir Mchergua), Ariana (Kalaat Landalous, Sidi Thabet), Manouba (Tebourba, Jdaida, Elbattan, BorjElaamri, Elmorneguia), Bizerte (Manzel Bourguiba, Mateur, Joumine), BenArous (Mhamdia, Mornègue), and Nabeul (Manzel Temime, Manzel Bouzelfa, Kelibia). The area surveyed and the geographical locations are mentioned in Figure 1. These regions represent four distinct bioclimatic levels: humid, sub-humid, upper semi-arid and middle semi-arid. Fields had a minimum area of 0.5 ha, and the minimum distance be-

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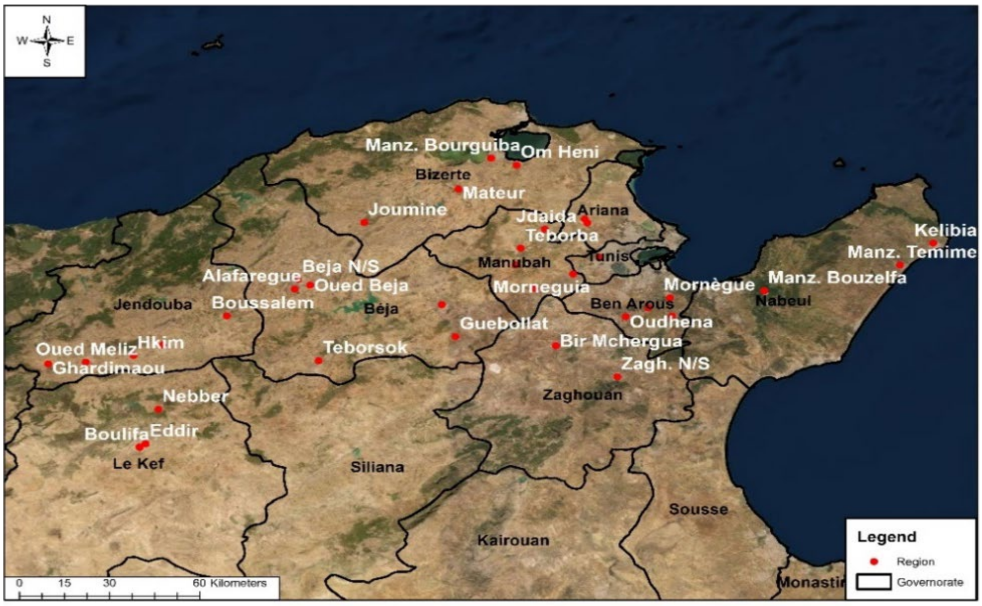


Figure 1. Map of visited regions during survey in northern Tunisia

tween fields was kept to 3–6 km. During the survey, each field's GPS position and region's climatic conditions were recorded (Table 1). The inspection protocol consists of collecting randomly 3–4 wheat plant samples located at nine points along the two diagonal transects of a wheat field, 30 plants in total. Wheat samples were put in paper bags and then transferred to the laboratory for further counting and analysis (Iram & Ahmad 2005). In addition, growers were asked for information on cropping system, cultivars, agricultural practices (sowing density, inputs, previous crop, fungicide treatment employed), and other observations about other diseases if they exist.

**Assessment of disease prevalence, incidence, and severity.** Global appreciation of tan spot disease presence in the visited regions allowed to calculate the prevalence as a simple ratio of the number of wheat fields showing foliar symptoms over the total visited fields:

$$\text{Prevalence (\%)} = \frac{\text{wheat fields showing foliar symptoms}}{\text{total of visited wheat fields}} \times 100 \quad (1)$$

The severity of each disease was determined using incidence and severity parameters (James 1971; Stubbs et al. 1986).

The incidence of disease was calculated as follows (Cooke 2006):

$$\text{Incidence (\%)} = \frac{\text{number of infected plants in the sampling}}{\text{total number of plants in the sampling unit}} \times 100 \quad (2)$$

Severity has been rated visually based on the estimation of the leaf area infected percentage of the three upper leaves, and conversing to score using a 0–5 scale, where 0: no symptoms, and 5: fully leaf dead, according to the findings of Bockus and Claassen (1992) and Raymond et al. (1985).

**Isolation and identification of *P. tritici-repentis*.** The symptomatic sampled leaves from visited fields were cut in fragments, disinfected in 3% hypochlorite solution for 3 min, rinsed twice in sterile distilled water, and placed in Petri dishes containing filter paper soaked water. To induce sporulation of the fungus on the leaf fragments, the Petri dishes were submitted to an alternation of fluorescent light and dark for 24 h to 19 °C (Lamari & Bernier 1989). After 2–3 alternation cycles, Petri dishes were observed under a binocular lens to verify the morphological characters of *P. tritici-repentis* based on Zillinsky (1983) description. If the presence of *P. tritici-repentis* was confirmed, a single spore was isolated from a leaf fragment with a steel needle and transferred to V8-PDA medium (agar 20 g, glucose 20 g, $CaCO_3$ 3 g, V8-juice 150 mL, 850 mL boiling potato). Single spore cultures were then incubated in the darkness at 20 °C for seven days (Lamari & Bernier 1989; Ali et al. 2010).

**Association with agricultural practices and environmental factors.** In order to highlight a potential association of tan spot with agricultural practices and environmental conditions, a logistic regression analysis was used (Yuen et al. 1996). To do this, incidence and severity data were classified into distinct groups of binomial qualitative data.

https://doi.org/10.17221/103/2022-PPS

Table 1. Characteristics features of surveyed wheat fields in several regions of northern Tunisia

| Region | Latitude and longitude | Bioclimatic stage | ACR (mm) |
|---|---|---|---|
| Ghardimaou | 36°26'54.52" 08°26'15.41" | humid | ≥ 800 |
| Joumine | 36°55'34.03" 09°23' | | |
| Oued Beja | 36°73'31" 9°21'38" | sub-humid | 600–800 |
| Alaferegue | 36°42'04.93" 09°10'30.95" | | |
| Kelibia | 36°83'99" 11°07'05" | | |
| Manz. Temime | 36°78'9" 10°95'39" | | |
| Manzel-Bourguiba | 37°08'35" 9°4'59" | | |
| Mateur | 37°02'19.99" 09°39'53.15" | | |
| Jendouba | 36°30'43.10" 08°46'48.9" | | |
| Mjez Elbab | 36°66'29" 9°24'52" | upper semi-arid | 500–600 |
| Teborsok | 36°27'34.01" 09°14'48.07" | | |
| Boussalem | 36°36'37.11" 08°58'21.27" | | |
| Teborba | 36°50'23.23" 09°51'05.61" | | |
| Elbattan | 36°79'25" 9°85'54" | upper semi-arid | 500–600 |
| Elmorneguia | 38°45'07.19" 10°00'28.08" | | |
| Mornegue | 36°40'17" 10°17'50" | | |
| Manzel Bouzelfa | 36°40' 10°34' | | |
| Kalaa-Andalous | 37°04'41" 10°10'89" | | |
| Sidi Thabet | 36°90'89' 10°05'53" | | |
| Borj Elamri | 38°42'07.49 09°53'40.89" | | |
| Beja N/S | 36°43'59.95" 09°11'03.72" | | |
| Bir Mcherga | 36°30'36" 09°57'23" | middle semi-arid | 400–500 |
| Kef-Eddir | 36°10'03" 08°42'36" | | |
| Boulifa | 36°09'59" 08°42'41" | | |
| Nibber | 36°17'42" 08°46'03" | | |

ACR – annual cumulative rainfall

Thus, the values ≤ 30% and > 30%; ≤ 10 and > 10 were chosen for disease incidence and severity, respectively, yielding a binary dependent variable. The independent variable variety was grouped into four classes according to their response (outcome). The independent variables included agricultural practices and bioclimatic conditions, were categorised, and were given in the analysis (Table 2). Contingency tables of disease intensity and the independent variables were built to represent the bivariate distribution of fields according to two classifications (e.g. district by tan spot incidence) and frequency (Table 3). The association of wheat tan spot intensities with agricultural practices and bioclimatic conditions was analysed using logistic regression (Yuen et al. 1996), with R procedure software (version 4.1.2). The logistic regression model allows to evaluate the importance of multiple independent variables that affect the response variable (Fininsa & Yuen 2001; Zewde et al. 2007; Hailemariam et al.

Table 2. Categorization of variables used in analysis for a survey of wheat tan spot disease in northern regions belonging in four bioclimatic stages

| Variable | Variable class | No. of fields |
|---|---|---|
| Bioclimatic stage | humid | 14 |
| | sub-humid | 45 |
| | upper semi-arid | 61 |
| | middle semi-arid | 14 |
| Variety | Maâli | 46 |
| | Karim | 48 |
| | Carioca | 3 |
| | Khiar | 11 |
| | Razzak | 8 |
| | Monastir | 12 |
| | Ismir | 4 |
| | Nasr | 2 |
| Sowing density | low (≤ 120 kg/ha) | 34 |
| | high (>120 kg/ha) | 100 |
| Previous crop | legumes | 25 |
| | cereals | 63 |
| | horticulture crops | 20 |
| | fallow | 15 |
| | industrial crops | 15 |
| Severity | ≤ 10% | 101 |
| | > 10% | 33 |
| Incidence | ≤ 30% | 107 |
| | > 30% | 27 |

Industrial crops – tobacco and fenugreek

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Table 3. Independent variable by disease contingency table for logistic regression analysis of wheat tan spot during the survey seasons in northern of Tunisia

| Variable | Variable class | Disease intensity (%) | | | |
|---|---|---|---|---|---|
| | | severity | | incidence | |
| | | ≤ 10 | > 10 | ≤ 30 | > 30 |
| Bioclimatic stage | humid | 99.25 | 0.74 | 98.50 | 1.49 |
| | sub-humid | 85.07 | 14.92 | 84.32 | 15.67 |
| | upper semi-arid | 94.02 | 5.97 | 93.28 | 6.71 |
| | middle semi-arid | 100.00 | 0.00 | 100.00 | 0.00 |
| Variety | Maâli, Razzak | 81.50 | 18.50 | 81.50 | 18.50 |
| | Karim, Carioca | 68.63 | 31.37 | 68.63 | 31.37 |
| | Khiar, Ismir | 86.66 | 13.33 | 86.66 | 13.33 |
| | Monastir, Nasr | 100.00 | 0.00 | 100.00 | 0.00 |
| Sowing density | ≤ 120 kg/ha | 99.25 | 0.74 | 99.25 | 0.74 |
| | > 120 kg/ha | 82.08 | 17.91 | 81.34 | 18.26 |
| Previous crop | legumes | 97.76 | 2.23 | 97.01 | 2.98 |
| | cereals | 89.55 | 10.44 | 88.80 | 11.19 |
| | horticulture crops | 98.50 | 1.50 | 97.76 | 2.23 |
| | fallow | 99.25 | 0.74 | 98.50 | 1.49 |
| | industrial crops | 94.02 | 5.97 | 93.28 | 6.71 |

Industrial crops – tobacco and fenugreek

2020). Logistic regression calculates the probability of a given binary outcome (response) as a function of the independent variables (McCullagh & Nelder 1989). If the probability of the outcome is denoted as $P$, the logistic regression model assumes that the logarithm of the odds of $P$ $[P/(1 - P)]$, which equals logit $P$, is a linear function of the independent (Yuen et al. 1996). In this study, the binary outcome was the probability that tan spot incidence exceeds 30% and severity exceeds 10% in a given wheat field. The importance of the independent variables on disease incidence or severity was tested first with each variable alone, then with all other independent variables. In the logistic regression model, maximum likelihood methods calculated parameter estimates. The deviance and the Chi-square value were analysed for the significant effect of the independent variables and variable classes on disease incidence and severity. The likelihood ratio test was tested against a Chi-square value (McCullagh & Nelder 1989).

## RESULTS

**Tan spot symptoms, distribution and prevalence.** During the survey, 200 wheat fields were visited in 28 cereal districts of Northern Tunisia. Tan spot disease was present in 67% of districts but with different incidence and severity intensities and various symptoms. Three types of foliar symptoms were observed: namely necrosis with chlorosis, necrosis, and brown spots with chlorosis. Necrosis with chlorosis symptoms was frequently detected (rate of 59.7%) in surveyed fields. Necrosis symptom was observed at the rate of 5.97% on leaves sampled in only Manzel Temime and Elbattan regions, while brown spots with chlorosis symptoms occurred in 34.33% of symptom cases. Means of recorded disease prevalence were high in some regions such as Oued Beja (87.5%), Manzel Bourguiba (83.3%), and Oued Mliz (72.5%), but were low in other regions like Bir Mchergua (25%), Guebollat, and Zaghouan north (30%) (Table 4). According to belonging regions to bioclimatic stages, the disease prevalence varied between different bioclimatic stages. The highest prevalence value was recorded in the sub-humid stage (63.87%). In the upper semi-arid stage, the prevalence was about 55.67%, reaching 45.25% in the humid stage. The lowest values were recorded in the middle semi-arid stage (30%) (Table 4). During the survey, weather conditions varied among bioclimatic stages and districts. At the sub-humid stage, the temperatures ranged from 8 to 25 °C during the wheat growth period, which is favourable to inoculum germination in field and tan spot disease development. In addition,

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Table 4. Prevalence of tan spot disease in distinct districts and bioclimatic stages during survey seasons

| Bioclim. stage | Region | Governorates | Visited fields | Prevalence (%) |
|---|---|---|---|---|
| Humid | Ghardimaou | Jendouba | 7 | 55.50 |
| | Joumine | Bizerte | 13 | 35.00 |
| | mean | – | – | 45.25 |
| Sub-humid | Oued Mliz | Jendouba | 10 | 72.50 |
| | Ouedbeja | Beja | 16 | 87.50 |
| | Mateur | Bizerte | 9 | 40.00 |
| | Manzel Br. | Bizerte | 11 | 83.30 |
| | Manzel Tem. | Nabeul | 9 | 66.60 |
| | Manzel Bou. | Nabeul | 6 | 75.00 |
| | Kelibia | Nabeul | 10 | 60.00 |
| | Mornègue | Ben Arous | 12 | 50.00 |
| | mean | – | – | 63.87 |
| Upper semi-arid | Ejdaida | Manouba | 14 | 78.50 |
| | Elbattan | Manouba | 14 | 80.00 |
| | Teborba | Manouba | 9 | 52.00 |
| | Borj./Elmr. | Manouba | 8 | 40.00 |
| | Kalaat Land. | Ariana | 13 | 61.00 |
| | Mhamdia | Ben Arous | 10 | 40.00 |
| | Nebbir | Kef | 6 | 55.00 |
| | Teborsok | Beja | 11 | 50.00 |
| | Mjez Elbab | Beja | 12 | 66.60 |
| | Bousalem | Jendouba | 9 | 40.00 |
| | Jendouba | Jendouba | 12 | 58.30 |
| | mean | – | – | 55.67 |
| Middle semi-arid | Kef - Eddir | Kef | 9 | 35.00 |
| | Zaghou. N/S | Zaghouan | 12 | 30.00 |
| | Bir Mcher. | Zaghouan | 10 | 25.00 |
| | Guebollat | Beja | 9 | 30.00 |
| | mean | – | – | 30.00 |

Bioclim. stage – bioclimatic stage; Manzel Br. – Manzel Bourguiba; Manzel Tem. – Manzel Temime; Manzel Bou. – Manzel Bourzelfa; Borj./Elmr. – Borj Elaamri/ Elmornaguia; Kalaat land. – Kalaat Landalous; Zaghou. N/S – Zaghouan North and South; Bir Mcher. – Bir Mchergua

rainfall and humidity varied belong districts and months. A highly significant difference in rainfall on disease development ($P$ = 0.000 016) and a significant difference in humidity ($P$ = 0.003 83). No significant effect of wind on tan spot progression ($P > \alpha = 0.05$).

**Tan spot incidence and severity.** Incidence and severity percentages of a tan spot on durum wheat for different variables were presented in Table 3. Results showed that 21.35% of visited fields have high disease incidence (> 30%), and 23.88% have high severity (> 10%), while the low incidence and low severity constituted the majority of recorded values in visited fields (Table 2). The disease incidence and severity varied along the bioclimatic stages. The highest incidence (> 30%) and the highest severity (> 10%) values were recorded in the sub-humid stage, where the rainfall ranged from 600 to 800 mm. The disease is less severe and has a decreased incidence in areas belonging to humid (≥ 800 mm) and middle semi-arid (400–500 mm) stages.

A low incidence was recorded in 98.5% of visited fields belonging to the humid stage, and 100% of fields located in the middle semi-arid. A low severity was observed in 99.25% of visited fields belonging to the humid stage, and 100% of fields located in the middle semi-arid.

The level of tan spot incidence and severity also varied among wheat varieties. The highest incidence and severity were recorded in fields planted with Maâli, Razzak, Karim, Khiar Carioca and Ismir varieties, while the lowest values were registered for Monastir and Nasr varieties (Table 3). The incidence and severity did not exceed 30% and 10%, respectively, for the sowing density inferior to 120 kg/ha, while these values increased with sowing density. Tan spot was observed in various wheat fields, which were planted in rotation with different crops such as cereals, legumes (bean, faba bean), horticulture crops (tomato, cucumber, and watermelon), industrial crops (tobacco, fenugreek) and fallow. The high disease incidence and severity values were recorded in wheat fields that were preceded by crop cereals and industrial crops such as tobacco but were low if the wheat was planted in rotation with fallow and market gardening (Table 3). Of the surveyed fields, 47% were continually planted with cereals crops.

During the survey, we observed variations in rainfall and humidity among the visited districts. The high incidence and severity were recorded during a high relative humidity of about 79.5 to 81% and cumulative monthly rainfall ranging from 53 to 95 mm during February and March.

**Association with agricultural practices and environmental factors.** The association of tan spot with the independent variables (variety, previous crop, bioclimatic stage, mean rainfall in April, mean relative humidity in May, mean temperature, mean wind, and sowing density) is presented

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Table 5. Independent variables used in logistic regression modeling intensity of wheat tan spot and likelihood ratio test (LRT) for five variables

| Independent variable | Degrees of freedom | Tan spot severity LRT | |
|---|---|---|---|
| | | Chi-squared | $P (> \chi^2)$ |
| Variety | 3 | 18.089 0 | 0.000 421 7 |
| Previous crop | 3 | 10.891 0 | 0.012 330 0 |
| Bioclimatic stage | 2 | 18.503 0 | 0.000 096 0 |
| Mean rainfall | 1 | 12.894 0 | 0.000 329 6 |
| Mean relative humidity | 1 | 2.299 6 | 0.021 790 0 |

in Table 5. Mean incidence and severity of tan spot were significantly correlated with bioclimatic stage ($\chi^2$ = 18.503, df = 2, $P$ = 0.000 096), variety ($\chi^2$ = 18.089, df = 3, $P$ = 0.000 421 7), previous crop ($\chi^2$ = 10.891, df = 3, $P$ = 0.012 33), mean rainfall ($\chi^2$ = 12.894, df = 1, $P$ = 0.000 32), and mean relative humidity ($\chi^2$ = 2.299, df = 1, $P$ = 0.021 79). Conversely, tan spot incidence and severity were not significantly associated with mean temperature, mean wind, and sowing density.

The variety of Ismir, followed by Karim and Monastir, were associated with a high probability of disease severity (66.7% and 43.3%, respectively). However, Nasr and Khiar, followed by Maâli and Razzek, were associated with the lowest probability of disease severity.

A group of three variables: district bioclimatic stage, varieties, and rainfall (April) significance, were tested in a reduced multiple variable model. Analysis of deviance (logarithms of odds ratio) for these variables added one by one to the reduced model showed the importance of each variable and variable class (Table 5). The parameter estimates, and standard error related to the severity of tan spot disease were also presented in Table 6.

Five variables: bioclimatic stage of the region, variety, mean rainfall of April, and mean relative humidity of May were highly associated with tan spot severity when entered as single variables into the model, and last with other variables into the model, while previous crop had lost significance (Table 5). Analysis of deviance for the variables, parameter estimates and their standard errors is given in Table 6. Low tan spot severity (≤ 10%) exhibited a high probability of association with the stages humid and middle semi-arid, varieties of Nasr and Mahmoudi, legume and fallow as prevising wheat crop, and low rainfall during growth stages (Table 6).

**Fungal isolation and identification of *P. tritici-repentis*.** After incubation, the examination of the symptomatic wheat leaves showed the development of transparent conidia clinging to a brown conidiophore emerging from the leaf surface. Based on the morphological characteristics of *P. tritici-repentis*, we observed under a stereo microscopic

Table 6. Analysis of deviance, natural logarithms of odds ratio and standard error of the selected independent variables analyzing tan spot severity

| Independent variable[a] | Degrees of freedom | Residual deviance[b] | LRT[c] | | Variable class | Estimate[d] | Standard error of the estimate | Odds ratio |
|---|---|---|---|---|---|---|---|---|
| | | | DR | $P (> \chi^2)$ | | | | |
| Intercept | 133 | – | 169.647 0 | | | 5.256 2 | 4.892 6 | 1.074 |
| Variety | 3 | 152 | 18.088 9 | 0.000 421 7 | Var2 | 0.566 0 | 0.566 2 | 0.130 |
| | | | | | Var3 | 3.132 2 | 1.885 6 | |
| | | | | | Var4 | –18.780 0 | 1 236.540 0 | |
| Rainfall | 1 | 133 | 18.512 9 | < 0.000 1 | rainfall | 1.712 6 | 0.418 3 | 4.094 |
| Bioclimatic stage | 2 | 106.252 | 26.792 5 | < 0.000 1 | sub-humid | 3.715 7 | 0.937 6 | 2.329 |
| | | | | | up. semi-a. | 1.696 6 | 0.818 0 | |
| MRH | 1 | 100.990 | 5.262 4 | 0.021 791 | MRH | –1.380 2 | 0.749 2 | –1.842 |
| Previous crop | 3 | 91.252 | 9.738 2 | 0.012 330 | cereals | 0.789 0 | 0.89 06 | 1.910 |
| | | | | | hort. crops | 0.432 0 | 0.966 6 | 0.447 |
| | | | | | fallow | –1.943 6 | 1.207 3 | –1.610 |

DR – deviance reduction; hort. crops – horticulture crops; MRH – mean relative humidity; $P$ – probability of a $\chi^2$ value exceeding the deviance; up. semi-a – upper semi-arid; Var2 – group of varieties karim and carioca; Var3 – group of varieties khiar and Ismir; Var4 – group of varieties Monastir and Nasr

[a]Independent variables added in to the reduced model; [b]unexplained variations after fitting the model; [c]likelihood ratio test; [d]estimates from the model with the independent variables added in to a reduced model

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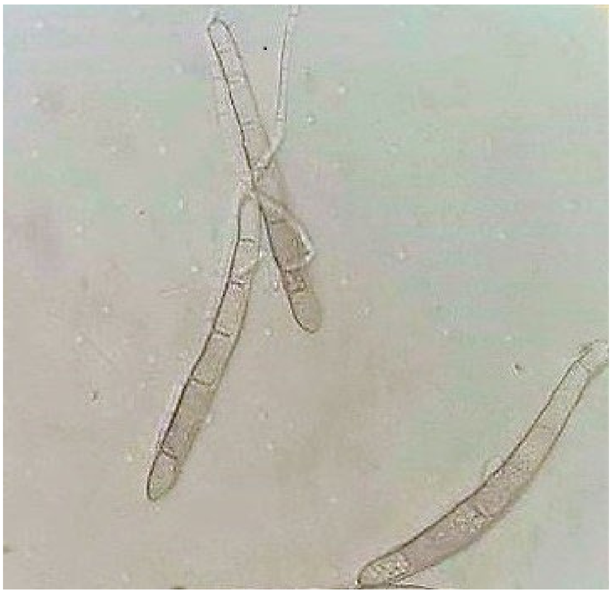

Figure 2. Microscopic observation of *Pyrenophora tritici-repentis* conidia

a typical form of conidia apex and septate and filiform conidia (Figure 2). Therefore, the observed infection in the fields from each sample collected was Tan spot disease (Figure 3). Ninety-five strains were isolated from several visited regions, with 23% detected in fungicide-treated fields. This could indicate the increasing adaptation of the fungus to the most used fungicides by Tunisian farmers.

## DISCUSSION

Several wheat fields belonging to four different bioclimatic stages of northern Tunisia were visited to detect tan spot and to determine incidence and severity. Bioclimatic conditions and agricultural practices were recorded during the investigation to assess their effect on the disease progression using logistic regression analysis. The wheat tan spot was widely spread in several districts with various incidence and severity rates. The regions of Oued Mliz, Oued Beja, Manzel Bourguiba and Manzel Bouzelfa belonging to the sub-humid stage were characterised by the higher prevalence degree and disease intensity. These high occurrences are in agreement with the results found during the spring season in surveyed Tunisian northern areas (Kamel & Cherif 2021; Tissaoui et al. 2022b). These areas are characterised by frequent and high precipitations (600–800 mm: sub-hid) and high humidity levels (72–86%) with tempered winter and spring ranging from 11 to 15 °C and 14 to 22 ℃, respectively. Such environmental conditions favour conidia germination of *P. tritici-repentis* (Lamari & Bernier 1994) and the development of tan spot disease (Kremneva & Gudoshnikova 2011). Indeed, it was established that tan spot progress increases in the areas with sufficient moisture, as in Russia (Kremneva & Gudoshnikova 2011) and India (Iram & Ahmad 2005). The association of weather factors and foliar disease development was confirmed in wheat strip rust (Sharma-Poudyal & Chen 2011), maize rust and leaf blight in Ethiopia (Fininsa & Yuen 2001), faba bean rust (Shifa et al. 2011), and garlic white rot (Zewde et al. 2007).

Figure 3. Tan spot typical symptoms in wheat

Statistical analysis showed that agricultural practices influence the disease's development and spread. The cereal monoculture and industrial crops (tobacco) as previous crop, increased the disease intensity compared to the case of wheat-fallow rotation. These results align with the interpretations of El Jarroudi et al. (2013), who found that monoculture and short rotations during intensification of wheat production and conservation tillage favour the tan spot development. In ad-

https://doi.org/10.17221/103/2022-PPS

dition, Fernandez et al. 2016 have established that crop rotations significantly influenced the severity of wheat leaf diseases in Western Canada. In other studies, Bankina et al. (2018) demonstrated that tan spot disease severity was higher in continuous wheat cropping compared to reduced soil tillage. However, severity decreases in the case of short rotations when fields are ploughed. Hence, the main source of *P. tritici-repentis* primary infection seems to be wheat residues where ascospores are developed and spread. Bailey et al. (2000) have also demonstrated that zero tillage was associated with more frequent isolation of *P. tritici-repentis* than with conventional tillage. In another work, Fernandez et al. (1998) concluded that the best rotations for a low level of wheat leaf spot infection leading to decompose wheat crop residues-were the case of two consecutive years of wheat followed by at least two years of noncereal crops, or by a noncereal crop and a summer fallow. In addition, the prevalent strains in Tunisia seem to exhibit new isolates resistant to the different chemical treatments used during the cropping season. Similar findings were reported by Colson et al. (2003) and Turner et al. (2021), highlighting the effect of fungicide treatment on increasing the resistance of strains.

Disease incidence and severity varied between the commercialised wheat varieties, showing distinct disease tolerance behaviour. The farmers grow high-producing varieties of durum wheat to satisfy national demand. However, these varieties were moderately susceptible to foliar disease (Anonymous 2016), such as septoria (Hassine et al. 2018). Particularly, recent studies revealed the susceptibility of varieties to tan spot in the juvenile stage, such as Maâli (Tissaoui et al. 2016), Karim and Razzek (Tissaoui et al. 2016; Kamel & Cherif 2021; Tissaoui et al. 2022b). The investigation showed variable reactions of commercialised wheat varieties to *P. tritici-repentis* which is in agreement with other studies about winter cultivars (Lamari & Bernier 1989b), Russian varieties (Šárová et al. 2002), and Tunisian varieties (Tissaoui et al. 2022a). The variety's response is due to existing resistance resources that are not still identified.

The conducted analysis indicates a clear association between bioclimatic conditions and agricultural practices with the development of wheat tan spot. The used regression model quantified the relative importance of variables representing how much the disease was increased or decreased as a function of the independent variables singly or in combination. This work allowed the detection of bioclimatic stage, varieties, previous crop, humidity relative, and rainfall as significant variables that influenced the incidence and severity of tan spot. It is in agreement with another work where the regression model demonstrated that the incidence and severity of wheat *Septoria tritici* blotch could be associated with agronomic conditions such as soil type, plant population, previous crop, and weed density (Hailemariam et al. 2020). The present survey showed that high tan spot incidence and severity were highly correlated with sub-humid stage, cereal as previous crop, high humidity, high rainfall, varieties Maâli, Razzak, Karim, Khiar Carioca, and Ismir. This showed that infection establishment and its spread on wheat depend on environmental and climatic conditions (Friskop & Liu 2021). The sub-humid region's high rainfall ensures the spores transport to the upper leaves and other neighbouring plants, and the humidity causes the reproduction of pathogens (Luck et al. 2011). In addition, some varieties seem susceptible to disease because they do not harbour race-specific resistance (Kokhmetova et al. 2017). In addition, the ability of virulent pathogens to overcome this resistance when varieties were used on a large scale (Yu et al. 2001). Therefore, their sowing in fields containing a high rate of pathogen inoculum due to lack of rotation promotes disease development (Bockus & Claassen 1992; Engle et al. 2006).

The analysis model was used in several researches to assess the association of environmental factors and agricultural practices with bean rust and common bacterial blight epidemics (Fininsa & Yuen 2001) and with white rot of garlic (Zewde et al. 2007). This model could also predict the risk of white mould disease on dry beans (Harikrishnan & Del Río 2008) and the potential loss of wheat yield caused by strip rust (Sharma-Poudyal & Chen 2011).

## CONCLUSION

Our investigation showed that the incidence and the severity of tan spot varied with bioclimatic stages, weather conditions, varieties, and cultural practices. High incidence and severity were closely correlated to susceptible varieties, high rainfall and humidity, and cereals as previous crop. The results suggest that assessed environmental conditions were relevant variables influencing wheat tan spot epidemics. Therefore, the suitable choice of varie-

https://doi.org/10.17221/103/2022-PPS

ties and agricultural practices contribute to reducing disease progression and minimising yield losses. These findings could be considered in developing an efficient tan spot management strategy.

**Acknowledgement:** We appreciate the INGC (Institut National des Grandes Cultures) technicians for the logistics of this study, and the staff of the regionals CTV for the contact with farmers during the visits carried out in nine governorates of northern Tunisia. We sincerely thank Dr. Walid Hamdane for the language revision.

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Recieved: September 19, 2022
Accepted: April 3, 2023
Published online: August 2, 2023