Effect of cereal cyst nematode *Heterodera filipjevi* on wheat yields in Turkey

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Abstract: The cereal cyst nematode *Heterodera filipjevi* (Madzhidov) Stelter is an important yield-limiting soil-borne pathogen of wheat- and barley-growing areas, particularly in the semiarid regions throughout the world. In Turkey, cyst nematodes have been detected in several localities of wheat- and barley-growing areas. This study was conducted to evaluate the effects of initial population densities (Pi) of *Heterodera filipjevi* on yield of susceptible (Seri-82) and resistant (Silverstar) wheat cultivars and to investigate nematode reproduction on wheat under rainfed conditions in Bolu Province in Turkey. The results indicated that cultivar Seri-82 showed higher sensitivity reaction to *H. filipjevi* than cultivar Silverstar. Yield reduction was negatively correlated with *H. filipjevi* initial population (Pi) for both wheat cultivars. Yield losses reached up to 40.5% and 8.54% for Seri-82 and Silverstar, respectively, at nematode density (Pi) (eggs + J2)/g soil of 44 and 38. There was a positive relationship between nematode Pi and final population (Pf), while the nematode reproduction factor (RF) was negatively correlated with nematode Pi of *H. filipjevi* on both wheat cultivars. Moreover, there was no relationship between the date of release of cultivars and their responses to the nematodes. Identification of resistance sources against *H. filipjevi* and the introduction of those into the selected germplasms by breeding programs is important for future studies.

Key words: Cyst nematode, *Heterodera filipjevi*, yield losses, wheat

1. Introduction

Wheat (*Triticum* spp.) and barley (*Hordeum vulgare* L.) are extensively grown under irrigated and nonirrigated conditions around the world due to their adaptability to wide climatic and geographic conditions as well as dietary traditions (Dababat et al., 2015). Abiotic and biotic agents are limiting factors to both wheat and barley cultivation, which negatively affect agronomic yield parameters such as plant height, straw yield, weight of 1000 kernels, and grain yield (Sikora, 1988; Scholz, 2001; Nicol et al., 2003; Dababat et al., 2016). Globally, plant parasitic nematodes (PPNs) are among the most commonly encountered soil-borne biotic agents that attack wheat and barley and cause significant yield loss of up to $100\text{ billion (Urwin et al., 1997; Bird and Kaloshian, 2003). Among the PPNs, the cereal cyst nematodes (CCNs) of the *Heterodera avenae* complex are important nematode pests of wheat, oats, and barley in many countries (Dababat et al., 2015). The CCN species *H. avenae* (Wollenweber), *H. filipjevi* Madzhidov (Stelter), and *H. latipons* (Franklin) are the most frequently encountered cyst nematode species attacking cereals, particularly wheat and barley (Rivoal and Cook, 1993; McDonald and Nicol, 2005). Economic losses in cereals caused by CCNs were documented and reported in several studies around the world (Rivoal and Cook, 1993; Peng et al., 2007; Subbotin et al., 2010; Mokrini et al., 2013; İmren et al., 2015; İmren et al., 2016; Mokrini et al., 2017; Fard et al., 2018) and were recently intensively reviewed by Dababat and Fourie (2018).

The CCN *H. filipjevi* sustains a global distribution and is probably the most damaging nematode in wheat- and barley-growing areas, particularly in semiarid areas where nematode damage increases under drought stress conditions (Rivoal and Cook, 1993; Nicol, 2002). Yield losses in wheat due to this CCN were reported to be between 10% and 40% in China, (Peng et al., 2007) and 40% and 92% in Saudi Arabia (Ibrahim et al., 1999). Smiley et al. (2005) reported a 35% yield loss in spring wheat in Oregon, USA, due to *H. filipjevi*, and more recently Fard et al. (2018) estimated yield losses in wheat yield ranging between 20% and 25% in Iran by the same nematode species. Also, Hajihassani et al. (2010) reported that grain yield loss caused by *H. filipjevi* occurred even at the lowest population density and reached a maximum loss of 48% with an initial population density (Pi) of 20 eggs and J2/ (g soil)⁻¹ in Iran.
In Turkey, the status, economic importance, and management of CCNs was reviewed by Dababat et al. (2014). Nevertheless, evidence is limited on the yield reduction of wheat or barley due to CCN species damage. There has been only one study to assess the impact of *H. filipjevi* on some wheat cultivars under field conditions in Ankara Province and the results indicated that there was a significant grain yield reduction (42%) in the studied wheat cultivars (Nicol et al., 2006). Similarly, the only study to evaluate the effect of *H. avenae* on six spring wheat cultivars under field conditions in Adana Province showed that there was a significant yield reduction (25.7%) (Imren and Elekcioğlu, 2014). Sahin et al. (2008) reported that *H. filipjevi* infests wheat and barley crops in several regions of Turkey where nematode population densities have reached 115 eggs and J2/g soil. Analyzing the relationship between the population density of CCNs and wheat yield is a crucial step in order to determine the degree of yield loss due to nematode infestation. However, limited information is available about the relationship between initial population (Pi) densities and yield response of wheat due to *H. filipjevi* existence in Turkish cereal production systems. Therefore, the objectives of this study were to estimate the negative impact of *H. filipjevi* on yield performance of two wheat cultivars with known reactions to *H. filipjevi* under rainfed conditions, and to study the effect of the Pi of *H. filipjevi* on wheat yield reduction of both cultivars.

2. Materials and methods

The effects of CCN *H. filipjevi* on the yield of 2 wheat cultivars [Seri-82 (susceptible) and Silverstar (resistant)] and the nematode reproduction factor (RF) were studied under field conditions over two consecutive growing seasons (2016–2017 and 2017–2018) in Çaydurt (40°45′32″N, 31°45′07″E), Bolu Province. Seeds were sown in September and the yield was harvested in July for both growing seasons. The experimental site was selected based on a previous survey conducted by Imren et al. (2016), who reported that the site was naturally infested with *H. filipjevi* at a rate of 344 cysts/kg soil. The climatic conditions of the experimental area are characterized by cold, snowy long winters and relatively short hot and dry summers. The annual temperature is averaged at 10.9 °C and precipitation is 573 mm with the majority of the precipitation falling in early winter (December) and in mid and late spring (April–May) (https://www.mgm.gov.tr/). The soil texture was clay-loamy with pH varying from slightly acidic to slightly alkaline.

Two wheat varieties, Seri-82 (susceptible to *H. filipjevi*) and Silverstar (resistant to *H. filipjevi*), were used in the experiments (Dababat et al., 2015). In order to compare the effect of nematode densities and to reach desired nematode population levels, half of the plots were treated with nematicide and the other half were left untreated as controls. The nematicide Nemathorin 10G, granules of 10% w/w fosthiazate (Syngenta International, Basel, Switzerland), was applied to the surface of the plots 1 day prior to sowing at a rate of 1.5 g/m² and then the top 15-cm soil layer of each plot was mixed thoroughly. Six rows of the treated and untreated plots were sown with the cultivars Seri-82 and Silverstar by hand at a density of 550–600 seeds/m². Each treatment (plot) was replicated 7 times (six rows 2 m wide × 5 m long). The experiments were arranged in randomized complete block design. Local management practices were applied in the trial throughout the growing seasons.

Ten soil subsamples were taken in a zigzag manner from each plot at a depth of 20–30 cm just before sowing for estimating the nematode Pi and at the end of the season for the final population (Pf) of *H. filipjevi*. Cysts extracted from 250 g of soil were recorded (Southey, 1986). Afterwards, cysts from each sample were smashed by using a tissue grinder in 50 mL of tap water and released eggs and J2s were counted under a stereomicroscope (V20, Zeiss, Jena, Germany). Pi and Pf were determined based on the numbers of eggs and J2/g soil, and RF (RF = Pf/Pi) was calculated accordingly (Scholz, 2001). At the end of the growing season (July) for both 2016–2017 and 2017–2018, all spikes from each plot were harvested with a small combine. Grain yield per plot was weighed and recorded. Average grain yield for each treatment was calculated and yield reduction percentage (%) was calculated based on the values derived from the treated and nontreated plots.

Dababat et al. (2016) reported 484 accessions with resistant reactions that were classified into one of five distinctive groups based on the RF: resistant (R) = RF equal to or less than 1; moderately resistant (MR) = RF between 1 and 2, a few more cysts than in a resistant group; moderately susceptible (MS) = RF between 2 and 3, distinctly more cysts than in a resistant group, but less than in a susceptible group; susceptible (S) = RF between 3 and 4, more cysts than in a susceptible group; and highly susceptible (HS) = RF above 4, cyst number higher than in a susceptible group and taking into account the reaction of the known control lines used in the study. Moreover, Smiley et al. (2004) described accession tolerance reactions by classifying them into 4 groups based on RF and yield potential. These groups were: tolerant (T) = good plant yield despite of high nematode attacks; moderately tolerant (MT) = plant yield moderate under nematode attack; intolerant (IT) = plants did not yield well even under low nematode pressure; and highly tolerant (HT) = plant yield good under high nematode attack.

The CCN data were normalized using the Shapiro–Wilk normality test before they were analyzed using analysis of variance (ANOVA) (Shapiro and Wilk, 1965).
Significant differences between lines were detected using the protected least significant difference at $P \leq 0.001$ using SPSS 17.0 (SPSS Inc., Chicago, IL, USA). Linear regression analyses were conducted to describe the relationship between the $H.\ filipjevi$ RF and grain yield for each line in the two experimental areas. Principal component analysis was used to determine population structure (Kendall correlation) using R 3.4.3 software to distinguish principal groups of wheat lines based on their tolerance to $H.\ filipjevi$. All other analyses (grain yield, RF, and yield loss) were performed using XLSTAT software 2016.02.28451 (Addinsoft, USA).

3. Results

The grain yields of the two cultivars were significantly higher ($P \leq 0.001$) in the fosthiazate-treated plots than those of untreated plots in both growing seasons, 2016–2017 and 2017–2018. Results indicated that $H.\ filipjevi$ caused significant yield reductions in the two cultivars, particularly in untreated plots. The reduction in yield was higher in the first season in 2016–2017 than in the second season in 2017–2018. In addition, the reduction in yield was obviously lower for the susceptible cultivar, Seri-82, than that for resistant cultivar Silverstar, as shown in Figure 1. The reduction in the grain yield of the two cultivars, Silverstar and Seri-82, ranged between 8.54% and 40.5% on average over the two growing seasons. The highest grain yield was 7700 kg/ha and was obtained from the nematicide-treated cultivar Silverstar (Figure 1).

In both growing seasons, the RFs of $H.\ filipjevi$ significantly varied among nematicide-treated and untreated plots. The average Pi of $H.\ filipjevi$ was estimated at 18 and 34 (eggs and J2) per gram of soil in 2016–2017 and 2017–2018, respectively. The results showed that the average RFs ranged from 0.67 to 6.83 in nematicide-treated and untreated plots, respectively. There was a significant negative correlation between the Pi of $H.\ filipjevi$ and yields of wheat cultivars ($y = -484.45x + 1125.6$, $r^2 = 0.6189$, $P \leq 0.001$), whereas a significant positive correlation between the Pi of $H.\ filipjevi$ and Pf was recorded (Figure 2).

The regression analyses of the combined data from both experiments showed inverse relationships between Pi density and wheat growth and yield parameters. These negative relationships were also described by linear models (Figures 3 and 4). The Pf increased with increasing Pi levels in both experiments, while the RF decreased (Figure 4). According to the changes in the Pi of $H.\ filipjevi$, reduction of yields of Silverstar and Seri-82 were determined as 8.54%, and 40.5%, respectively. Generally, the regression analyses showed that grain yield of Seri-82 was negatively correlated with Pi ($y = -116.71x + 814.39$, $r^2 = 0.5712$, $P \leq 0.001$) (Figure 3). Similarly, a negative response to Pi was found for grain yield of Silverstar ($y = -392.45x + 1030.6$, $r^2 = 0.7169$, $P \leq 0.001$) (Figure 4). Pf was positively correlated with Pi for Seri-82 and Silverstar, respectively (Figures 3 and 4). However, RF was negatively correlated with Pi for Seri-82 and Silverstar (Figures 3 and 4).

4. Discussion

The results of this study indicated that $H.\ filipjevi$ has great potential to cause damage and yield losses in wheat cultivation in Bolu Province, Turkey, where $H.\ filipjevi$-caused yield loss in cultivars Seri-82 and Silverstar averaged 8.54% and 40.5% in two-year experiments.
Figure 2. Principal component analysis (Kendall type) showing the population structure for a set of Silverstar and Ser-82 replications based on their tolerance reaction in field conditions.

Figure 3. Log-linear regressions of grain yield of Seri-82 and Heterodera filipjevi RF.
respectively. Likewise, Smiley et al. (2005) reported that nematicide application increased the yield of spring wheat by 24% in a field moderately infested by CCNs in southwestern Oregon, USA. Fard et al. (2018) conducted field trials to evaluate the impact of *H. filipjevi* on three wheat cultivars and their results showed significant reductions in grain yield ranging between 19.5% and 27.8%. Moreover, Hajihassani et al. (2010) conducted a microplot experiment to investigate the effects of *H. filipjevi* on the wheat yield of cultivar Sardari with different Pi values. They reported that nematode density of 2.5 eggs and J2/g soil caused yield reduction of 48%. Similarly, the damage caused by *H. filipjevi* to different wheat, barley, and triticale cultivars indicated significant reduction in grain yield by 52% (40%–73%) in field conditions in Khuzestan Province, Iran (Ahmadi et al. 2013). Also, the CCN species *H. avenae* caused significant yield loss in different wheat cultivars, varying from 4.3% to 25.7% in Adana Province of Turkey (Imren and Elekcioğlu, 2014). This paper revealed the negative impact of *H. filipjevi* on two wheat cultivars in field conditions, which is in agreement with the findings of these previous studies.

This study reports the first quantifiable evidence of a correlation between yield reduction and nematode density of *H. filipjevi* on wheat cultivars in Bolu Province, Turkey. The reproduction rates of *H. filipjevi* were in the range of 0.67–2.2 in the nematicide-treated plots and 2–6.8 in the untreated plots, suggesting that the fosthiazate application sustained an obvious suppressive effect on the nematodes in the experimental plots. Thus, the results of the study support the effect of nematicide treatment in wheat yield improvement and this is in agreement with other studies by Smiley et al. (2004, 2005). The obtained results indicated that yield could be increased when resistant or moderately resistant cultivars and seed treatments are combined. Also, the results showed that treating susceptible cultivars can be economically important in the reduction of nematode damage.

The results of this study also indicated an inverse relationship between the yield parameters and the RF in plots fosthiazate-treated and nontreated with the highest and lowest reproduction rates, respectively (Figures 3 and 4). The negative relationship between the Pi of *H. filipjevi* and yield loss of these two cultivars was supported by microplot trials of *H. filipjevi* infestation on wheat cultivar Sardari (Hajihasani et al., 2010). Smiley et al. (2005) evaluated wheat germplasm in both *H. avenae*- and *H. filipjevi*-infested fields and found that there was a reduction in yield as the nematode Pi increased. More recently, a yield loss study with *H. filipjevi* was conducted under field conditions by Fard et al. (2018) and the results were in line with our findings.

In conclusion, the present study indicated that *H. filipjevi* sustains a high potential to cause yield loss in Bolu’s wheat-growing areas of Turkey. This devastating nematode can spread fast and control measures such as resistant varieties, proper cultural practices, and seed treatment should be involved.

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