



# Distribution, population dynamics and damage potential of barley stem gall midge, *Mayetiola hordei* (Diptera: Cecidomyiidae) on cultivated barley in two semi-arid areas of North Tunisia

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## ARTICLE INFO

### Keywords:

*Mayetiola hordei*  
Biology  
Ecology  
Generations  
Infestation  
Sowing date

## ABSTRACT

Barley stem gall midge, *Mayetiola hordei* (Kieffer) (Diptera: Cecidomyiidae) is among the most destructive pests of barley (*Hordeum vulgare* L.) in the Mediterranean region of southern Europe and northern Africa. In Tunisia, the biology and ecology of this pest are not well documented. In addition to a management study, a study of *M. hordei* distribution, frequency of occurrence and population dynamics in barley crops was conducted in two semi-arid regions of North Tunisia. The pest was detected in 84% of all sampled fields across two years. Zaghouan region was identified as a high spot region with economic infestations registered in 67 and 56% of surveyed fields in 2013 and 2014, respectively. Three annual generations occurred at both sites, with two complete and one incomplete generation. The second generation (winter generation) is the most important one in terms of crop damage. The study showed that damage from this pest affects host plant growth at different developmental stages. The highest infestation was observed during tillering and elongation. The impact of different sowing date of barley on *M. hordei* infestation was also conducted. The infestation rate of barley by *M. hordei* increased by delaying sowing date. We demonstrate that *M. hordei* is a serious biosecurity threat to Tunisian barley's production and industry.

## 1. Introduction

In Tunisia, barley *Hordeum vulgare* L. is among the major crops and the second most widely cultivated cereal after durum wheat. This crop is cultivated on more than 30% of the total cereal arable land in Tunisia (El Felah, 2011), and is mostly sown in northern parts of the country where 52% of the total area cultivated in barley is planted (Ministry of Agriculture, 2014). More than 80% of barley is used for animal feed, and the rest is used for food and malt (El Felah and Medimagh, 2005). Barley is attacked each year by a large number of pests that depreciate yield and limit the production potential. In Tunisia, aphids and gall midges are the main described barley pests (Ben Fekih et al., 2013; Harbaoui et al., 2008; Bouktila et al., 2009).

The barley stem gall midge, *Mayetiola hordei* (Kieffer) (Diptera: Cecidomyiidae) is one of the most destructive pests of barley in most cereal Mediterranean regions of southern Europe and northern Africa (Gagne et al., 1991; Lhaloui et al., 1988; Lhaoui et al., 1992; Solh and Saxena, 2011). Recurrent droughts and favorable winter temperatures are the main factors causing *M. hordei* outbreaks (Solh and Saxena,

2011). This insect is capable of rapid and widespread population occurrence. The barley stem gall midge is a galling pest able to control and redirect plant development (Shorthouse et al., 2005), have high infestation rates and cause heavy yield losses. A damaged *M. hordei* field is characterized by the death or uneven growth of plants (Malipatil, 2008). All damage to seedling barley induced by *M. hordei* is the result of feeding by larval instars. Larvae feed at the base of the plant between the leaf sheath and the stem. Infestation by this pest on barley causes distinct pea-shaped swellings at the base of the stem (Gagne et al., 1991; Parker et al., 2001).

To manage cereal gall midges from the genus *Mayetiola*, several control measures are commonly used. The most practical methods are preemptive including the application of insecticides and planting date adjustment (Buntin and Bruckner, 1990; Buntin et al., 1990, 1992; Buntin, 1992; Ratcliffe and Hatchett, 1997; Morgan et al., 2005; Lhaloui et al., 2006). The best planting date management recommends sowing the crop after adult gall midge activity has stopped (Whitworth et al., 2010). However, the most effective and economical way to control these insects is the use of resistant cultivars (Ratcliffe and

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Hatchett, 1997; El Bouhssini et al., 1992; Lhaloui, 1995; Lhaloui et al., 1996, 2000; Williams et al., 2003). A strict gene-for-gene resistance relationship was demonstrated between some gall midges and their host plants. In fact, most gall-inducing insects are highly specific to their host plants and organs, and only induce galls on a single or closely related host species (Dreger-Jauffret and Shorthouse, 1992; Floate et al., 1996). Barley is the preferred host of *M. hordei* (Gagne et al., 1991). This insect has also been reported on wheat, oat and rye.

Despite the importance of the barley stem gall midge, limited information is available regarding its pest status, direct and indirect damage, and impacts on yield. This study is the first rigorous survey of the pest status, distribution and frequency of occurrence of *M. hordei* in five regions of Northern Tunisia. We focused on its population dynamics and importance of damage in the two most infested regions, Zaghouan and Kef, in 2013 and 2014. Secondly, a trial to assess the effect of different sowing dates as a cultural control method was performed.

## 2. Material and methods

### 2.1. Survey on distribution and frequency of occurrence of *M. hordei* in north Tunisia

A two-year survey (cropping seasons 2012–2013 and 2013–2014) was carried out over five regions of northern Tunisia corresponding to two bioclimatic stages: sub-humid (Bizerte and Béja) and semi-arid (Zaghouan, Siliana and Kef) in order to determine the distribution of barley gall midge infestations, their frequency of occurrence, the differences in levels of infestation and incidence observed between regions, and the intra and inter-annual variations between infestations and incidence levels.

Sampling was conducted across 50 barley fields in each region. Sampled fields were between 5 and 7 ha in size. Widely dispersed farms (5 km at least between fields) were selected in an attempt to represent different production conditions. All surveyed fields were cultivated by barley variety Manel and free from any insecticide application. At barley maturity, during the month of May, 50 plants were uprooted from each field and placed in a plastic bag for laboratory examination. At this time, *M. hordei* has completed all its generations thus no new infestations can be observed on barley plants. For each plant, all tillers were examined by peeling each leaf back to its node and the number of infested tillers was recorded. The number of larvae and pupae on all tillers was also summed in order to determine the percentage of infestation and incidence rate of *M. hordei* in each region. Infestation percentage was expressed as the number of infested tillers divided by the total number of tillers. The incidence rate was calculated as the number of flies per tiller divided by the total number of infested tillers. Moreover, the procedure described by Lafever et al. (1980) was adopted to monitor fields with economic levels of infestation (more than 20% of tillers infested) and fields with severe levels of infestation (more than 50% of tillers infested).

### 2.2. Population dynamics, damage to barley, and control of *M. hordei*

Field experiments were carried out in two barley fields (5 ha each) located in the two most infested regions of northern Tunisia: Zaghouan and Kef. In both regions, barley was sown on 28 November 2012 and 25 November 2013, which is around the conventional barley sowing date in Tunisia. Normal agricultural practices were followed and no insecticides were applied. Population dynamics, effects and control of *M. hordei* were assessed on barley cv Manel (Pedigree: L527/5/As54/Tra//2\* Cer/TolI/3/Avt/TolI//Bz/4/Vt/Pro//TolI ICB81-607-1Kf-1Bj-12Bj-11Bj-1Bj-1Bj-0Bj), which is one of the most cultivated varieties in Tunisia.

#### 2.2.1. Population dynamics of *M. hordei* in two semi-arid regions of north Tunisia

Population dynamics of *M. hordei* were studied for two successive cropping seasons (2012–2013 and 2013–2014) in each of the two barley fields in Zaghouan and Kef regions. Weekly sampling was conducted from September to August of each year. Two procedures were adopted: (i) For immature stages (eggs, larvae and pupae), 50 plants with symptomatic infestation (stunted plants with short, large, bluish-green color leaves) were sampled weekly from each field. Eggs, 1st, 2nd and 3rd instar larvae and pupae on all tillers were counted. (ii) For adult flies, ten 180° sweeps using a sweep net (diameter: 30 cm) were performed during early morning, in each field at each sampling date. After barley harvest, sampling was made on stubble and volunteer barley plants following the two procedures described above.

The number of eggs on leaves and the number of larvae and pupae at nodes or at the base of plants were counted using a binocular microscope. Larvae and pupae were exposed by pulling back the leaf or stem sheaths at the base or nodes. The pupal exuviae, without any remains, were recorded as exited by an adult of *M. hordei*. *M. hordei* population dynamics, the number of annual generations and the importance of population size between generations were established and compared between regions, seasons and years.

#### 2.2.2. Effect of *M. hordei* feeding on barley biomass

The biomass parameters were plant height, tiller number and shoot dry weight (Honsdorf et al., 2014). Effects of *M. hordei* on barley biomass parameters were determined for two successive cropping seasons in 2012–2013 and 2013–2014 in each of the two barley fields.

*M. hordei* damage was assessed for four developmental stages of barley: seedling, tillering, elongation and heading, by comparing plant height, tiller number and shoot dry weight between infested and non-infested plants. To assess the insect damage on barley biomass parameters, the following procedure was adopted: at each developmental stage, samples of 50 infested and 50 non-infested plants were uprooted. This step was repeated 4 times in each of the two barley fields (4 replicates). Plants were taken on diagonal transects at a rate of 1 plant every 10–15 m. For each plant category (infested and non-infested), plant height was measured using a ruler and the total number of tillers/plant was recorded. Fresh biomass was oven dried at 70 °C for 48 h to determine its dry weight. For all parameters, comparisons were made between infested and non-infested plants each year at each developmental stage.

#### 2.2.3. Field trials based on different sowing dates for managing *M. hordei*

The experimental design was a randomized block with three treatments. Each treatment corresponded to a different date of sowing barley (25 November, 13 December 2013 and 3 January 2014), arranged randomly within each of the four replicates. Every plot was composed of 6 rows with a length of 5 m and row spacing of 0.2 m. Normal agricultural practices were followed and a hand weeding was done when needed. No pesticides were applied. At the end of the cropping season, during the month of May, 50 plants were uprooted from each plot. The number of infested tillers and pupae per plant were recorded in order to measure infestation and incidence rates.

### 2.3. Statistical analysis

Data were expressed as mean  $\pm$  SD. For the regional survey, for each cropping season, infestation and incidence rates of *M. hordei* were compared between regions using one-way ANOVA followed by Duncan test with regions as fixed effect and fields as random effect. Inter-annual variations of infestations and incidences, in each region, were evaluated by t-test.

For the effect of *M. hordei* feeding on barley biomass, comparisons of infested and non-infested plants were made for each parameter at each development barley stage using t-tests.

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