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THE OBSERVATIONS ON THE DAMAGE OF *Cameraria ohridella* (Lepidoptera: Gracillariidae) FEEDING ON HORSE-CHESTNUT TREES (*Aesculus hippocastanum* L.) IN ESKİŞEHİR

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ABSTRACT: This paper gives insights into the behavior of *Cameraria ohridella* (Lepidoptera: Gracillariidae) at a regional scale on the basis of observations conducted in Eskişehir, Turkiye, from 2023–2024. Horse-chestnut trees (*Aesculus hippocastanum* L.) were randomly selected and the mined leaves were collected to determine the extent of the damage. Among the inspected locations, the infestation was observed only on the trees in Eskişehir Forest Nursery, while others were pest free. This could be due to (1) *C. ohridella* not having had time to reach other regions, (2) the establishment of the pest being inhibited by various biological and environmental factors, and (3) the role of leaf litter in the spread of *C. ohridella* in these areas. The results emphasize the importance of removing fallen leaves as a control measure to minimize the damage caused by *C. ohridella*. To the best of our knowledge, this is the first record of *C. ohridella* in Anatolia.

Keywords: Leaf-miner moth, pest distribution, pest control, urban ecosystem, Anatolia

ESKİŞEHİR'DE AT KESTANESİ (*Aesculus hippocastanum* L.) AĞAÇLARINDA BESLENEN *Cameraria ohridella* (Lepidoptera: Gracillariidae)'NIN HASARINA İLİŞKİN GÖZLEMLER

ÖZET: Bu makale, 2023-2024 yılları arasında Eskişehir, Türkiye'de yapılan gözlemlere dayanarak *Cameraria ohridella*'nın bölgesel ölçekteki davranışına ilişkin bilgiler vermektedir. At kestanesi ağaçları (*Aesculus hippocastanum* L.) rastgele seçilmiş ve zararın boyutunu belirlemek için galerili yapraklar toplanmıştır. İncelenen yerler arasında, sadece Eskişehir

Orman Fidanlığındaki ağaçlarda istila gözlenirken diğerlerinde zararlıya rastlanmamıştır. Bunun nedeni (1) *C. ohridella*'nın diğer bölgelere henüz ulaşmamış olması, (2) zararlının yerleşmesinin çeşitli biyolojik ve çevresel faktörler tarafından engellenmesi ve (3) *C. ohridella*'nın yayılmasında yaprak döküntüsünün rolü olabilir. Sonuçlar, *C. ohridella* popülasyonlarını yönetmek ve zararı en aza indirmek için bir kontrol önlemi olarak yaprak döküntülerinin temizlenmesinin önemini vurgulamaktadır. Bilgilerimize göre, bu *C. ohridella*'nın Anadolu'daki ilk kaydıdır.

Anahtar Kelimeler: Yaprak galeri güvesi, zararlı yayılışı, zararlı kontrolü, kent ekosistemi, Anadolu

INTRODUCTION

Some horse-chestnut trees (*Aesculus hippocastanum* L.), which have brown leaves and premature defoliation, draw an autumnal picture much earlier than usual. This phenomenon may be due to infestation by larvae of a tiny moth, the horse-chestnut leaf miner *Cameraria ohridella*. *C. ohridella* is an invasive pest that was first recorded in Lake Ohrid, Macedonia, in 1985 and subsequently described as a new species (Deschka & Dimić, 1986). It has rapidly spread and become a problem throughout Europe over the past 30 years. For example, because of the damage the pest cause, Germany has ceased using horse-chestnut trees for ornamental purposes (Reinhardt et al., 2003). DNA analyses indicate that the pest originates from the Balkans (Valade et al., 2009). The pest was first recorded in Turkiye in 2004 in a recreational forest called the Belgrad Forest in Istanbul (Cebeci & Sabiha, 2007). However, to date, there is limited information on its distribution across Turkiye.

The species is host-specific and feeds mainly on *Aesculus* spp., most significantly white flowering horse-chestnuts (*Aesculus hippocastanum* L.). The pest does not target sweet chestnut (*Castanea sativa*) but has been observed in *Acer* and *Fraxinus* species (Kopačka & Zemek, 2017). However, the larvae do not complete their life cycle because of saponins that are present in the leaves of these species (Kopačka & Zemek, 2017). Saponins are the seconder metabolite products which defense the plants against insects and microorganisms by inhibiting larval development (Küçükkurt & Fidan, 2008). Once the eggs hatched, the larvae of *C. ohridella* feed between the upper and lower epidermis of the leaves. The pest can cause extensive damage by laying up to 70 eggs on horse-chestnut leaves, with multiple overlapping generations possible in a year under favorable climatic conditions (Ivinskis & Rimsaite, 2006). For instance, in mediterranean countries, the moth can have up to five generations but no more than three generations in northern countries (Gilbert et al., 2005). Larvae undergo five developmental stages and the pupae overwinter inside the fallen leaves (Krivosheina & Ozerova, 2020).

Although *C. ohridella* does not cause mortality by damaging the leaves, it damages the mechanism of photosynthesis, converting sunlight into sugar. This may affect the reproduction of horse-chestnut trees and increase their susceptibility to other disturbances (Krivosheina & Ozerova, 2020). Since horse-chestnut trees are used ornamentally in parks, gardens, and roadsides in cities, the real impact of defoliation is mostly aesthetic. Summer browning and premature leaf fall can cause public concern, as trees lose their majestic appearance and may appear to be dying.

There is no obvious solution for controlling *C. ohridella*. Control measures include chemical, biological, and cultural approaches. Chemical application is not recommended because it may harm pollinators and beneficial insects. The pest has natural enemies such as parasitoids and entomopathogens; however, their incidence is low, and they are poorly synchronized with the life cycle of the pest (Ivinskis & Rimsaite, 2006; Volter et al., 2022). Birds are more effective than arthropods but can only consume pupae inside fallen leaves at the end of the season, which is usually not enough to control the population of *C. ohridella*. While the cutting of infected trees is the definitive solution, the best short-term approach is to remove and burn fallen leaves at the end of the seasons (Pavan et al., 2003). This method kills pupae before they overwinter and emerge as adults, causing more damage following year (Pavan et al., 2003). It is affordable and feasible especially for small-scale managements. Long-term solution is to breed resistant horse-chestnut trees (Gubka et al., 2024).

The aim of the present study is to provide information about the scope of damage at the regional scale and the distribution of *C. ohridella* in Turkiye.

MATERIALS AND METHODS

This observational study was conducted in the years, 2023-2024, in Eskişehir (39° 45' 58.2948" N, 30° 31' 36.1704" E), a city located in the Central Anatolia Region of Turkiye, where horse-chestnut trees are commonly used in parks, gardens, and urban roadsides. Eskişehir has an arid climate with dry summers and cold winters at an elevation of 800 m.

To assess the presence and the extent of the damage caused by *C. ohridella*, we carried out field observations. For this reason, we visited parks, gardens, Eskişehir Forest Nursery and urban roadsides of Eskişehir to look for main symptoms of the pest; browning and mined leaves. The samples were collected from the places where the symptoms were observed, which was only in the nursery. In the nursery, one group of horse-chestnut trees was mixed with scots pine in a garden setting and others were on the roadside. From each of the two groups, ten leaves from five trees were randomly collected approximately 2m in height. The leaves were sealed in plastic bags and taken to the laboratory to obtain the adults for imaging. Since the infestation is not severe and the larvae caused individual mines, we counted the mines on the collected leaves to assess the extent of the damage.

RESULTS AND DISCUSSION

Identification of Cameraria ohridella

The inspections revealed that the damage on the leaves was caused by *C. ohridella* (Figure 1). The leaves had mines with pupae inside and some had dead larvae in the mines. The pupae were light brown in color and almost 4 mm long (Figure 1a). June, July and August generations were recorded in 2024. The adults' emergence was observed at the beginning of August. The adult was brown in color with white stripes and was approximately 4 mm long (Figure 1d). This is the first study that the invasive pest feeding on horse-chestnut trees was found inner Anatolia in Turkiye.

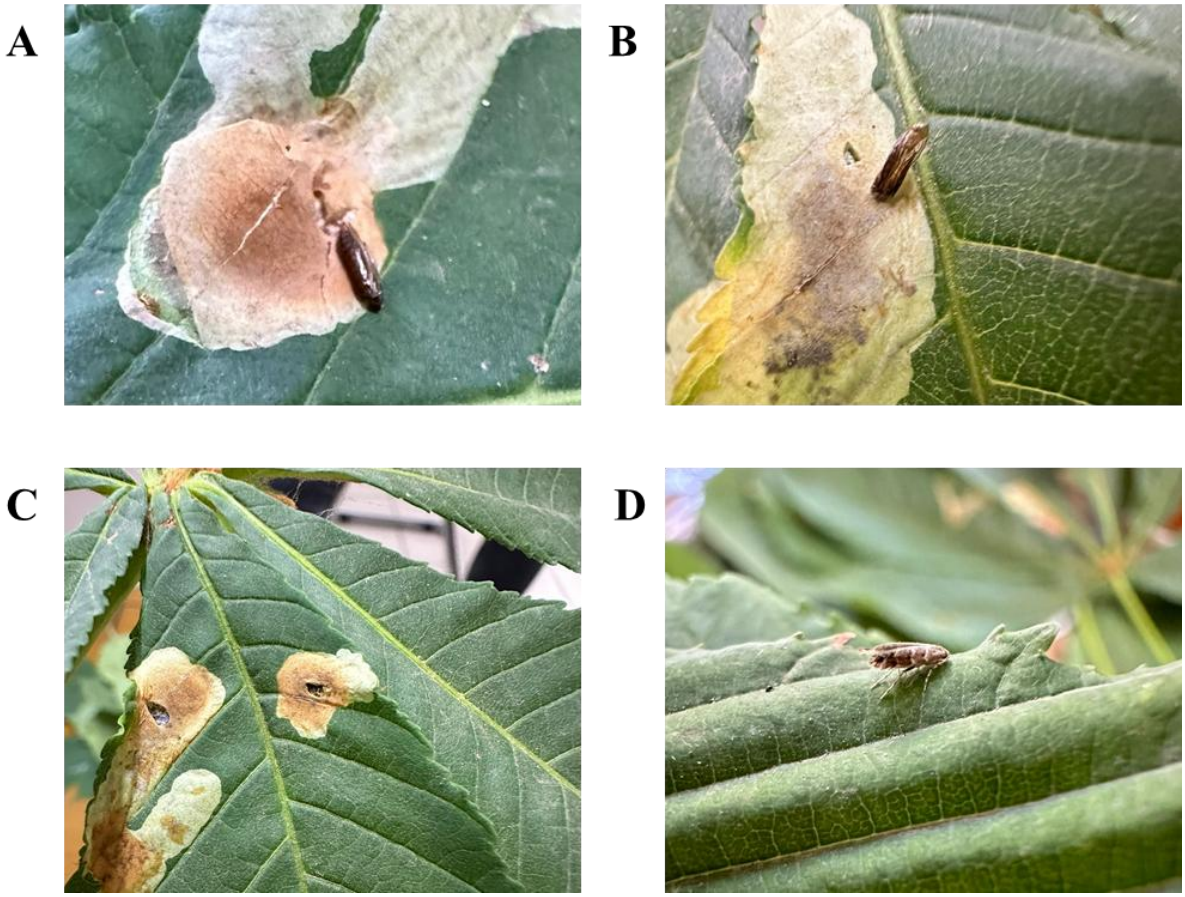

Figure 1. The pupae of *Cameraria ohridella* (A), emerging an immature adult of *Cameraria ohridella* from the mine (B), the exit holes after the adults have emerged (C), and the adult of *Cameriaria ohridella* (D).

Infestations Differ Locally

The level of the damage differed by location inspected. The number of mines on the leaves of the horse-chestnut trees was higher in the nursery than other places. The damage did not occur at the whole-tree level but occurred mostly in the lower branches of the trees. This could be because *C. ohridella* has a low population density, which has not yet caused significant damage.

The trees in the garden were more infested than the trees used for roadside greenery (Figure 2). The average number of mines was almost ten times greater for the trees in the garden setting than for those used as roadside planting (Figure 2). In the garden, the horse-chestnut trees were planted close to each other and had leaf litter underneath, which was not observed for the trees used for roadside planting. The better to clean leaf litter is the better to manage the pest because leaf litter serves as a resource for the next generations of *C. ohridella*. The pupae overwinter in the leaves from the previous year, leading to increased damage in the following season. For example, research on the effectiveness of leaf removal under urban conditions revealed that the complete removal of both dead litter and pupae resulted in a significant decrease in infestation; however, the rate of the infestation increased significantly where dead litter remained (Pavan et al., 2003). Thus, complete removal of litter before adult emergence is highly recommended, marking it as the best practice (Krivosheina & Ozerova, 2020; Pavan et al., 2003).

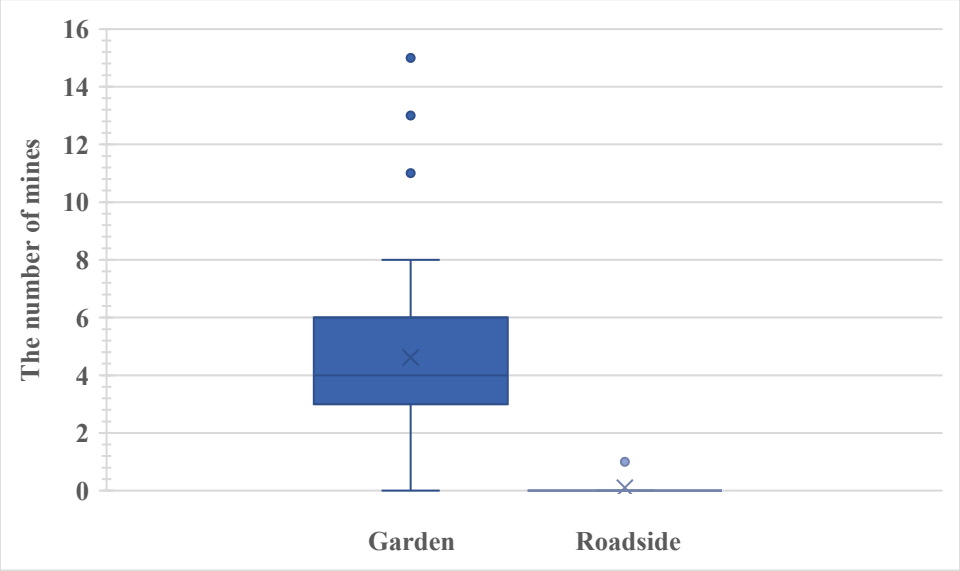

Figure 2. The number of mines caused by *Cameraria ohridella* on the leaves of horse-chestnut trees used for garden and roadside planting in the nursery.

Although some studies indicate that *C. ohridella* damage is more severe in city centers where trees are likely to be stressed but this was not the case in Eskişehir region. In gardens and parks in downtown, the trees had no mined leaves but had rusty brown blotches bordered with a clear yellow band, which is the symptoms of horse chestnut leaf blotch, a fungal disease caused by *Guignardia aesculi* (Peck) V.B. Stewart (Botryosphaeriales: Botryosphaeriaceae). Although the interaction between the pest and the fungus has not yet been fully understood, *G. aesculi* could be the superior rival of *C. ohridella* (Kopačka et al., 2021). Thus, the establishment of the pest might be hindered by a variety of biological and environmental factors.

Other possible explanations for the presence of the pest only in nursery is that (1) *C. ohridella* might be initially established in the nursery and might not have had sufficient time to reach the city center and (2) having an active train rail crossing in the nursery might contribute in the spread of *C. ohridella.* It is the fact that the pest spreads by flight within cities but causes damage over large areas through passive transportation (i.e., wind, cars, and trains) (Gilbert et al., 2004). Indeed, initial observations of the pest were often made in areas such as vehicle parking lots (Augustin et al., 2009).

The horse-chestnut seedlings distributed throughout the city have been produced from the seed sources within the nursery. Given the widespread use of horse-chestnut trees in parks, gardens, and forests, the pest is expected to spread further with horse-chestnut seedlings (Krivosheina & Ozerova, 2020). Currently, no damage caused by *C. ohridella* was observed in the horse-chestnut seedlings.

Changes in climate could also stress horse-chestnut trees, increasing their susceptibility to the pest damage and reducing their ability to recover. Rising temperatures and changing precipitation patterns may create more favorable conditions for the pest, enabling it to expand its range and increase its population density, consequently the damage. As a result, regions previously unaffected by the pest may become vulnerable, necessitating increased monitoring and management efforts to mitigate its impact on urban and natural landscapes. For example, in Lithuania, horse-chestnut leaf miners spread through 39 cities in only 4 years, and their

habitat range is expected to expand in the future (Ivinskis & Rimsaite, 2006). The climate for Eskişehir region was projected to be hotter and drier in 50 years by an approximately 2°C increase in temperature (Basciftci et al., 2021). Therefore, understanding and addressing the impacts of climate change on the distribution and behavior of *C. ohridella* is crucial for developing effective management strategies to protect horse-chestnut trees in the future.

CONCLUSION

This study provides crucial insights into the presence and damage of *Cameraria ohridella* in Eskisehir, Turkiye, marking the first record of this pest on the Anatolian side of the country. The infestation was higher in the garden settings which has more leaf litter than in roadside plantings, highlighting the role of leaf litter in sustaining *C. ohridella* populations. Effective management practices, such as the removal and burning of fallen leaves, are essential for controlling the spread and impact of this pest. Given the potential for further spread facilitated by transportation and climatic conditions, ongoing monitoring and implementation of control measures are necessary to protect horse-chestnut trees from significant aesthetic and physiological damage. This study also emphasizes the need for more extensive research on the distribution and impact of *C. ohridella* across Turkiye to develop comprehensive management strategies. Further studies should focus on (1) extending the research to other regions of Turkiye, (2) evaluating the effectiveness of different control measures under varying conditions, (3) identifying fungal associations with the pest, and (4) investigating the impact of climate change on the pest's distribution and behavior.

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AUTHOR CONTRIBUTIONS

Zeynep ÇİÇEKLİ: Conceptualization, Methodology, Data curation, Formal analysis, Writing - original draft. **Hakan DEMİRBAĞ:** Formal Analysis, Investigation, Data curation. **Saliha VOYVOT:** Supervision, Writing-review, Editing.

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CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

ETHICS COMMITTEE APPROVAL

This study does not require any ethics committee approval.

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