

REVIEW

The box tree moth *Cydalima perspectalis*: a review of biology, invasiveness, management practices and future perspectives of control strategy in Europe

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Abstract

The box tree moth, *Cydalima perspectalis* (Lepidoptera: Crambidae), is a significant invasive pest threatening boxwood (*Buxus* spp.) in Europe, North America, and parts of Asia. Since its initial detection in Europe in 2006, *C. perspectalis* has spread rapidly, causing widespread damage to both ornamental and wild boxwood populations. Although extensive investigations have been conducted on its biology, reproduction, ecology, and phenology, achieving fully sustainable control strategies in Europe remains challenging, even with numerous studies and pest management efforts documented in the literature. It is a highly polyvoltine species, with larvae that aggressively consume boxwood foliage leading to defoliation and plant death. The economic impact in Europe has been particularly severe in natural landscapes, especially in historical gardens. *C. perspectalis* is highly adapted to feeding on boxwood. It is plausible that the microbiome of larvae might detoxify phytochemicals and modify plant defense thus facilitating their survival and proliferation. This review consolidates the current knowledge on *C. perspectalis*, including its biology, origin, and distribution. Based on currently available literature, effective management strategies, which primarily rely on monitoring and early detection, are discussed. Due to the challenges in controlling this pest and the lack of effective natural enemies an integrated pest management (IPM) approach is recommended. This strategy combines biological, chemical, and mechanical methods to reduce populations and limit their destructive impact. Furthermore, the pest's ability to neutralize the natural toxins in boxwood leaves increases the risk of insecticide resistance development. Consequently, understanding the microbial interactions between *C. perspectalis* and its host plant could offer further pest control strategies by targeting the microbiome to disrupt the detoxification process, making the insect more susceptible to boxwood defenses.

Keywords: box tree moth, defoliation, invasiveness, microbiome, pest species

Introduction

Globalization, through the expansion of trade, increased human mobility, and environmental changes facilitates the spread and establishment of invasive species beyond their native geographical ranges. International plant trade has significantly increased the frequency of invasive species introductions, posing serious threats to local ecosystems and agriculture. One of the most problematic species is the box tree moth

(BTM), *Cydalima perspectalis* (Walker, 1859), which has become a significant threat to the health and survival of various boxwood species (*Buxus* spp.), widely recognized for their high ornamental value in gardens and parks. The native range of the BTM encompasses the subtropical regions of Southeast Asia, with the earliest reports of its presence in China, dating back to the mid-19th century, followed by reports from India. By

the early 21st century, this species had expanded into Europe, where it rapidly spread across new territories.

Effective management of the box tree moth requires a thorough assessment of the efficacy of existing control methods, considering its biology, reproductive capacity, and susceptibility to various treatments. This paper presents a comprehensive review of the current knowledge of biology, dispersal mechanisms, and the damage inflicted by the box tree moth, to improve pest management strategies. The primary objective is to enhance understanding of this issue and explore practical solutions with a particular focus on natural enemies and biological control, to aid in monitoring and controlling the pest population while minimizing negative impacts on the natural environment and protecting boxwood plants. Given the increasing challenges associated with environmental protection and maintaining ecosystem balance, it is crucial to develop effective yet sustainable pest management methods, including those targeting box tree moths. Based on the available literature and global research, with a particular emphasis on European studies, this work summarized a management strategy for combating this invasive pest on boxwood plants.

Biological characterization of *Cydalima perspectalis*

Taxonomy

Cydalima perspectalis, the box tree moth, is a species classified in the family Crambidae, first identified and described as *Phakellura perspectalis* by the English entomologist Francis Walker in 1859 (Mally and Nuss 2010). It has been variably classified under the spilomeline genera *Palpita*, *Diaphania*, and *Glyphodes*. However, no convincing reasons have been cited for its placement in these genera. In 2008, Streltsov identified key morphological distinctions between *C. perspectalis*, *Diaphania*, and *Glyphodes*, leading to the establishment of a new monotypic genus, *Neoglyphodes* (Streltsov 2008). However, this classification was not thoroughly examined within the broader context of the Spilomelinae subfamily, which encompasses 277 genera and over 3,700 species, highlighting the need for further taxonomic and phylogenetic revision (Mally and Nuss 2010). To address this issue, a recent comprehensive analysis of the mitochondrial genome (mitogenome) has been conducted by Gao *et al.* (2023) to investigate the phylogenetic relationships of *Cydalima perspectalis* across different taxonomic levels. Their findings revealed that *C. perspectalis* shares a closer evolutionary relationship with *Pygospila tyres* within the Spilomelinae subfamily than with other members

of the Crambidae and Pyraloidea superfamilies (Gao *et al.* 2023).

Invasion and geographic range in the new area

Cydalima perspectalis is a species native to Southeast Asia, particularly the subtropical regions of China and India. The first reports of its presence in China date back to the mid-19th century. In the early 21st century, this species was introduced into Europe, rapidly spreading across the continent. The initial European appearance of the BTM was reported in 2006 in Baden-Württemberg, southeastern Germany, where it was accidentally introduced through boxwood plants imported from Asia (Bras *et al.* 2019; Krüger 2008; Mally and Nuss 2010). Considering the significant damage it caused, it is believed that the moth could have been introduced into Europe up to 2 years earlier (Billen 2007; Krüger 2008). In 2007, its presence was subsequently reported in Switzerland (Billen 2007) and the Netherlands (Mally and Nuss 2010). By 2020, the moth had spread across almost all of Europe (Bras *et al.* 2019), with its range extending from northern regions such as the United Kingdom (Mitchell 2009; Salisbury *et al.* 2012; Plant *et al.* 2019), Lithuania (Paulavičiute and Mikalauskas 2018), Russia, and Ukraine (Budashkin 2016) to southern areas including Hungary (Sáfián and Horváth 2011), Croatia (Koren and Črne 2012), Bosnia and Herzegovina (Ostojić *et al.* 2015), Montenegro (Hrnčić and Radonjić 2017), Slovakia (Bakay and Kollár 2018), Malta (Agius 2018), Italy (Raineri and Mariotti 2017), Serbia (Stojanović *et al.* 2015), Greece (Strachinis *et al.* 2015), and Portugal (Vieira 2020). The moth has also been observed in Turkey (Hizal *et al.* 2012), Georgia (Matsiakh *et al.* 2018), and Iran (Ghavidel *et al.* 2021; Zamani *et al.* 2018). Additionally, in 2018, the first occurrence was reported in North Africa – in Algeria (Haddad *et al.* 2020). Around the same time, the moth was recorded in North America (Coyle *et al.* 2022), where it is noteworthy that it remains under strict official control (EPPO 2024).

Studies analyzing the genetic diversity of European populations of *C. perspectalis*, based on mitochondrial DNA sequences of cytochrome oxidase subunits I and II, identified two haplotypes – HTA and HTB – out of 12 known from Chinese populations, indicating multiple introductions into Europe from eastern China (Bras *et al.* 2018; Matošević *et al.* 2017). The rapid spread of the BTM in Europe has been primarily caused by globalization, inadequate regulation of the ornamental plant trade, and the fact that it is not classified as a quarantine pest by the European and Mediterranean Plant Protection Organization (EPPO), leaving it without official control measures. The current

distribution of box tree moth aligns with the climatic model developed by Nacambo *et al.* (2014), which predicted the gradual expansion of this species' range in Europe, extending as far as the Fennoscandian Peninsula and northern Scotland, excluding high-altitude areas, where climatic conditions act as natural barriers to its spread (Kenis *et al.* 2013; Nacambo *et al.* 2014).

In Poland, Bereś *et al.* (2021) conducted detailed monitoring that documented the 8-year spread of the BTM. The species was first detected in 2012 near Michałków, in the Sowie Mountains (Blaik *et al.* 2016). Over the subsequent 6 years, it caused extensive damage across various regions (Bury *et al.* 2017). Initially spreading from the south, the moth moved west to east, gradually expanding northward. By the end of 2020, it had been observed across the entire country, with the highest population densities recorded in southern Poland (Bereś *et al.* 2021).

Morphological characterization and life cycle

Box tree moth adults exhibit two distinct color morphs in their adult stage: a light form with white wings edged in brown and characteristic white discoidal spots and a less common melanistic form with light brown wings displaying a purple gloss and a contrasting white crescent-shaped discoidal spot on the forewings (Blaik *et al.* 2016; Dobrzański *et al.* 2018) (Fig. 1).



Fig. 1. Two distinct color morphs of *Cydalima perspectalis*: a light (left) and characteristic melanistic form (right)

In Hungary, researchers identified three morphological forms: two light and one dark (Tuba *et al.* 2015). In turn, studies conducted by Bereś *et al.* between 2020 and 2022 revealed that Polish populations, collected in Warsaw, Rzeszów, and Wrocław, exhibited only one light and one dark form (Bereś *et al.* 2024). These moths have a wingspan of approximately 3.5-4 cm, making them the largest species among the European representatives of the Crambidae family (Blaik *et al.* 2016).

Adult moths possess a sucking proboscis adapted for nectar feeding (Mally and Nuss 2010; Matsiakh *et al.* 2018). Female moths lay clusters of 5-20 trans-

parent eggs on the underside of boxwood leaves, deep within the bush, which complicates monitoring efforts (Bereś *et al.* 2021). The eggs are round, approximately 1 mm in diameter, initially, light yellow and covered with a transparent secretion that helps them adhere to the leaves. As the eggs develop, they enter the "black head" stage, where the black head capsules of the developing larvae become visible through the eggshells (Bereś 2019) (Fig. 2). After a few days, the larvae hatch. The larval stage consists of 6-7 instars (Capelli *et al.* 2018). Newly hatched larvae are yellow about 1.5-2 mm long, with black, shiny heads equipped with chewing mouth parts (Fig. 2). Initially, light green with brown lines, the larvae darken as they grow, developing characteristic black stripes with white dots, light bristles along their sides, and two rows of black warts on their dorsal side (Leuthardt and Baur 2013; Matošević 2013; Dobrzański *et al.*; 2018 Bereś 2019) (Fig. 2). The pupa reaches a length of 1.5 to 2 cm and is concealed within a cocoon of white silk, located between boxwood leaves and twigs. Initially green with dark stripes, the pupa gradually turns brown as it develops, with a segmented pattern resembling the brown coloration found on the moth's wings (Fig. 2) (Maruyama *et al.* 1993; Leuthardt and Baur 2013; Matošević 2013).

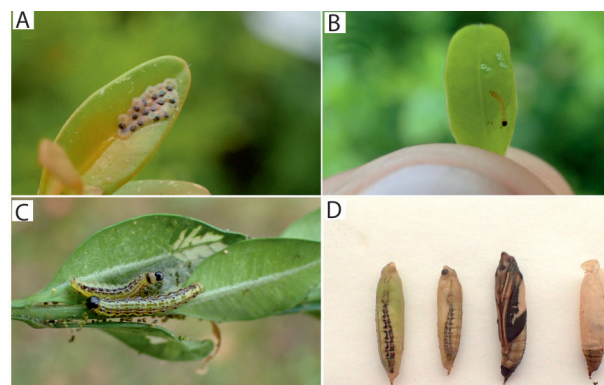


Fig. 2. The developmental stages of *Cydalima perspectalis*.

Panel a – egg cluster, b – young larva, c – older larvae (3rd instar larvae), d – pupae (color change from young pupa to empty pupa)

Understanding the developmental cycle of the box tree moth is crucial for effectively planning and implementing control strategies. Despite nearly two decades of invasion in Central Europe, this species' biology remains a significant focus of scientific research (Bereś *et al.* 2022). Previous studies reported that the box tree moth could have one or two generations per year in Europe (Nacambo *et al.* 2014). However, because of climate change, recent observations suggest that this species can now produce as many as three to four full

generations each year, depending on local climatic conditions. The minimum temperature required for moth development ranges from 8°C to 12°C (Bereś *et al.* 2021). Remarkably, in certain regions, such as China, up to five generations per year have been reported (Wan *et al.* 2014). The box tree moth has a high reproductive potential, with a single female capable of laying up to 500 eggs (Mally and Nuss 2010; Leuthardt *et al.* 2013; Bereś *et al.* 2021). Meteorological conditions have a strong influence on the development of box tree moths. Laboratory tests carried out by Farahani *et al.* (2021) at a constant temperature (25°C) indicate the duration of the individual development stages of the pest (Tab. 1).

Table 1. Mean duration of life stages and intervals of *Cydalima perspectalis* under laboratory conditions at 25°C

Different life stage/period (day)	Duration stage in laboratory conditions in days (in 25°C)	
	minimum	maximum
Incubation period	4	6
1st Instar larval period	2	5
2nd Instar larval period	2	7
3rd Instar larval period	2	4
4th Instar larval period	2	5
5th Instar larval period	3	6
6th Instar larval period	5	12
Pre-pupal period	1	2
Total larval period	20	31
Pupal period	6	8
Developmental time	33	44
Female longevity	5	27
Male longevity	4	25
Life span	39	62

Larvae typically hatch 4-5 days after the eggs are laid, initially feeding in groups before dispersing throughout the boxwood shrubs until they pupate (Bereś *et al.* 2021). The developmental cycle includes 6-7 larval instars, with each stage lasting approximately 3 days at a constant temperature of 25°C (Bereś *et al.* 2021). After 3-4 weeks, the larvae reach their full size of about 4 cm, at which point they enter the pupal stage. Before this occurs, larvae hide deep in the shrub, surrounding themselves with a dense cocoon reinforced with silk, which complicates their mechanical removal and provides protection from both low temperatures and pesticides (Dobrzański *et al.* 2018). The pupal stage usually lasts for 10 days at 25°C. Studies showed that a temperature increase of just 1.5°C can reduce

this period by 2 days. Adult moths have an average lifespan of about 2 weeks (Korycinska and Eyre 2011; Leuthardt and Baur 2013). A simplified diagram of the pest's development cycle is presented in Fig. 3. The life cycle of the box tree moth is temperature-dependent,

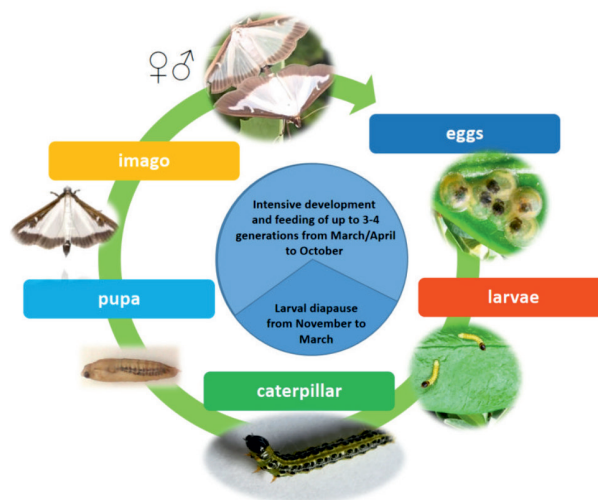


Fig. 3. The scheme of life cycle of *Cydalima perspectalis*

with development duration varying according to environmental conditions. At 20°C, the development from egg to adult moth takes approximately 40 days. During warm summer months, the life cycle accelerates, facilitating rapid population growth. Seasonal adaptations, such as reduced temperature sensitivity and accelerated development, have been observed, influencing the phenological variability and synchronization of the BTM across different habitats (Matsiakh *et al.* 2018). In autumn, as temperatures decrease, the species enters diapause. In Europe, the third instar larvae of the second generation typically enter diapause. Research indicates that the larvae become more sensitive to photoperiods during this period. A shortening day length and food scarcity caused by the intense feeding of larvae from both generations can also trigger diapause (Wan *et al.* 2014; López and Eizaguirre 2019). During this period, the moth can survive the winter in its larval stage. Nacambo *et al.* determined that a day length of 13.5 hours is critical for diapause induction, which lasts at least 8 weeks (López and Eizaguirre 2019; Nacambo *et al.* 2014). During this time, overwintering larvae cover themselves in thick silk, forming characteristic cocoons between the leaves deep within the shrub, which allows them to withstand temperatures as low as -30°C (Korycinska and Eyre 2011; Leuthardt *et al.* 2013; Nacambo *et al.* 2014; Bereś *et al.* 2021).

Understanding these characteristics is crucial for developing effective management strategies to reduce

the damage caused by this invasive pest. Consequently, controlling the box tree moth population requires continuous monitoring and analysis of its life cycle, along with timely interventions to protect boxwood plants.

Economic importance

Host and harmfulness

For centuries boxwood has played a significant role in garden design, both in historic parks and private gardens. Initially favored in monastic and palace gardens for its ornamental quality and ability to be shaped, boxwood's dense, evergreen foliage and high tolerance for heavy pruning have made it indispensable for creating hedges, topiaries, and borders. This plant adds elegance and structure to garden compositions.

The genus *Buxus*, belonging to the Buxaceae family, comprises approximately 90 species that naturally occur in most tropical regions as well as in the Mediterranean area (Leuthardt and Baur 2013). Boxwoods are rarely consumed due to the presence of highly toxic compounds, which can only be accumulated and metabolized by specialized herbivorous species. Moreover, a recently performed comprehensive metabolomics study by Hay *et al.* (2024) provides the first metabolome depiction of *Buxus* response to a BTM invasion, manifesting a significant increase of triterpenoid and steroidal alkaloid synthesis. However, their significance and ecological functions in defense against herbivores need detailed studies. Insects use different strategies for overcoming these barriers, including co-opting of metabolic activities from microbial associates (Wielkopolan and Obrepalska-Stepłowska 2016; Mason *et al.* 2019; Višnovská *et al.* 2020). The importance of microbial communities in herbivores and their relationship with plant hosts has been extensively studied in Coleoptera (Wielkopolan and Obrepalska-Stepłowska 2016; Wielkopolan *et al.* 2024) and some Lepidoptera (Mason *et al.* 2019). Interestingly, previous research has shown that the larvae of the BTM accumulate dibasic alkaloids from their host plants, while monobasic alkaloids are metabolized and/or excreted. Notably, no traces of these compounds have been detected in adult moths (Leuthardt *et al.* 2013). However, the data regarding the mechanism of detoxification remains unknown.

In Europe, box tree moth larvae feed on all native species and cultivated varieties in parks and gardens (Leuthardt and Baur 2013; Nacambo *et al.* 2014; Wan *et al.* 2014). According to the European and Mediterranean Plant Protection Organization (EPPO), the primary boxwood species serving as host plants for *C. perspectalis* include *Buxus balearica*, *B. microphylla*, *B. sempervirens* and *B. sinica*.

In Asia, larvae have also been reported feeding on other plants, including *Euonymus* species (*E. alatus* and *E. japonicus*) and *Ilex purpurea* (Mally and Nuss 2010). In Poland, particularly in the southeastern region, a few larvae have also been observed feeding on *Euonymus* species, *Cotoneaster* species, *I. purpurea*, *Prunus laurocerasus* (cherry laurel) and *Murraya paniculata* (orange jasmine) (Bereś 2019; Dhakal and Ad-desso 2023).

Initially, young larvae cause “windowing” damage by consuming tissue from the surface of the leaves. As they mature, larvae consume entire leaf blades, leaving only the veins and edges, which often curl characteristically (Fig. 4). Scientific observations have shown that in the absence of leaves, the late larval stages can also feed on shoots, stripping the bark (phloem), disrupting nutrient and water transport, and significantly weakening the plant (Lemic *et al.* 2023).

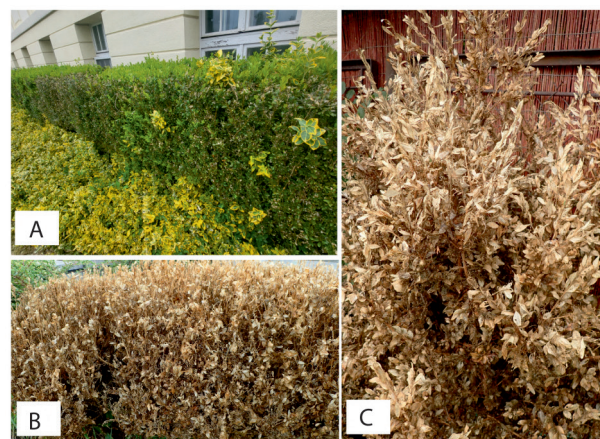


Fig. 4. Symptoms of boxwood damage caused by *Cydalima perspectalis*.

Initial damage (panel A), and complete defoliation with boxwood shrubs entirely stripped of leaves due to pest feeding (panels B and C)

An interesting study by Kulfan *et al.* (2020) described the spatial distribution of larvae feeding on shrubs throughout the growing season. It was observed that in spring, most larvae feed on the lower parts of the shrubs. During summer, they move to the middle sections, and by autumn, they feed on the upper leaves. Intense feeding results in extensive defoliation covered with silk webs, leading to the plant's inability to perform photosynthesis, which ultimately causes its death (Kulfan *et al.* 2020).

In Central Europe, BTM typically develops 2-3 generations per year, depending on weather conditions, as was mentioned above (Plant *et al.* 2019). In Poland, the box tree moth easily survives in the winter, and in

spring, as temperatures rise, the overwintering larvae begin feeding, with the first adult moths emerging by late April (Bereś 2019). Additionally, high temperatures, long photoperiods, and the species' high fertility accelerate the development of subsequent generations, increasing the number of generations per year and thereby enhancing the pest's harmful impact. Notably, scientific observations indicate that in 2018, due to the hot and dry summer in southeastern Poland, a fourth generation of the pest likely developed, since active moths were identified in November (Bereś 2019).

Box tree moths can disperse over distances of up to 10 km per year (Korycinska and Eyre 2011). Long-distance invasions are facilitated by the international transport of boxwood seedlings, while local spread is supported by monocultural plantings in gardens, such as hedges, as well as plant nurseries and online sales. Consequently, due to extensive feeding by caterpillars and the rapid spread of the species, boxwood has been struggling against this aggressive "Asian invader" for years. As a result, gardeners, landscape architects, and amateur horticulturists are increasingly abandoning the cultivation and design of boxwood compositions in home gardens, urban settings, and parks due to the damage inflicted by this pest. Moreover, the challenges and costs associated with pest management are prompting plant nursery owners to reduce the propagation of boxwood.

Current management methods

Cultural methods

In Europe, *C. perspectalis* lacks natural predators capable of significantly controlling its population, necessitating the use of mechanical, biological, or chemical control methods. Furthermore, due to the overlapping nature of successive generations, pest management must be sustained throughout the entire growing season.

Agricultural practices for managing BTM primarily focus on inspecting shrubs and mechanically removing feeding larvae. This labor-intensive approach is effective when damage to shrubs is limited, particularly in spring after larvae emerge from diapause, as it significantly reduces population density. Amateur gardeners can employ practical techniques such as placing mats under shrubs and shaking plants to dislodge larvae, which should be promptly destroyed through crushing, burying, or burning. It is important to note that caterpillars produce silk threads, allowing them to cling to lower branches during this process. Additionally, gardening guides recommend washing off caterpillars with a strong jet of water, such as from a pressure washer, which can damage the delicate bodies of the larvae without harming the boxwood.

A supportive mechanical protection strategy against the boxwood moth also includes the use of light and pheromone traps. In 2007 Kawazu *et al.* identified three active components (Z)-11-hexadecenal (Z11-16: Ald), (E)-11-hexadecenal (E11-16: Ald), and (Z)-11-hexadecenol (Z11-16: OH) that can act as female sex pheromones. Several commercial options are available that utilize synthetic substances to attract male moths, which can help reduce further reproduction (Bereś *et al.* 2022; Kazerani *et al.* 2023; Plant *et al.* 2019; Santi *et al.* 2015). However, according to the literature pheromone traps are more effective for monitoring the spread of this pest in new areas and for determining the appropriate timing for control measures, rather than for direct efficient control (Barbero *et al.* 2024; Plant *et al.* 2019; Santi *et al.* 2015). In turn, light traps attract moths of both sexes.

The analysis of volatile compounds in larval excrement performed by Molnár *et al.* (2017) yielded intriguing findings, identifying substances such as guaiacol, (\pm)-linalool, and veratrole, all of which displayed repellent properties. When a synthetic blend of these compounds was applied to boxwood plants, a marked reduction in the number of eggs laid was observed, confirming the blend's efficacy as an oviposition deterrent, and providing further insights into behavioral responses relevant to pest management strategies.

The development of plant varieties with enhanced resistance to diseases and pests can significantly reduce the need for pesticides and other plant protection products, ultimately benefiting the environment. However, current recommendations for managing the box tree moth do not include effective breeding methods. Various boxwood cultivars exhibit phenotypic differences. Traits such as leaf color, shape, and hardness may influence female preferences for oviposition sites. Furthermore, variations in the chemical composition of leaves, particularly alkaloid content, may serve as chemical or physical cues for females, impacting their behavior and/or larval development (Leuthardt and Baur 2013).

Cultivation of disease-resistant plant varieties as a strategy for pest management

Research on commonly cultivated evergreen boxwood species, such as *Buxus sempervirens* and its cultivars 'Sempervirens,' 'Rotundifolia,' 'Argenteovariegata,' and 'Aureovariegata,' as well as *B. microphylla* 'Faulkner,' has revealed that female moths exhibit specific preferences for both oviposition and feeding. In particular, 'Rotundifolia' and 'Aureovariegata' are favored for egg-laying and feeding over other varieties (Leuthardt and Baur 2013; Leuthardt *et al.* 2013; Kholghahmadi *et al.* 2023). In 2023, Kholghahmadi *et al.* published a study examining the influence of two boxwood species,

B. hyrkana and *B. microphylla*, on the ecophysiology of this invasive pest. The findings revealed differences in developmental time, fecundity, and nutritional indices of the pest depending on the boxwood species. Although the moth can develop on both species, it reproduces significantly more efficiently on *B. hyrkana* than on *B. microphylla*. Nutritional indices were also notably higher in *B. hyrkana*. Although the nutrient content in the leaves of both species was similar, the larvae and adult of *C. perspectalis* differed in protein content, triglyceride levels, and alkaline phosphatase activity, potentially affecting the pest's reproductive capacity. Collectively, these factors contribute to the higher fecundity of BTM reared on *B. hyrkana*, which in turn underscores the importance of host plant quality in determining the reproductive parameters of this pest. Although chemical, visual, or tactile cues typically guide these preferences, the exact mechanisms of pest-plant interactions in the case of the box tree moth remain unexplored (Sugio and Plantés 2020).

Chemical control

The chemical control of the BTM involves the application of synthetic insecticides, which must be used in strict accordance with the manufacturer's guidelines, as well as environmental and public health regulations. To mitigate the risk of resistance development in the pest population, it is crucial to rotate insecticides with different active ingredients. Furthermore, many of these products can be harmful to beneficial insects, so their use should be limited to situations where other methods have failed.

The number of registered active substances for the control of box tree moths may vary in individual countries. In some countries, the following active substances have been registered: chlorantraniliprole, methoxyfenozide, abamectin, emamectin benzoate, thiacloprid, trichlorfon, chlorpyrifos, alpha-cyhalothrin, beta-cyhalothrin, cypermethrin, spinosad and tau-fluvalinate (Qian XiaoLan *et al.* 2018; Somsai *et al.* 2019; Stan and Mitrea 2020).

In Poland, acetamiprid-containing (neonicotinoid), tebufenozid (diacylohydrazine), emamectin benzoate-based (macrocyclic lactone), spinosad (spinozyne) are the only chemical substances registered for BTM control.

In addition to confirming the toxicity of chemicals to the box tree moth, it is crucial to prevent the development of resistance to the applied substances. This presents a significant challenge for plant protection strategies aimed at minimizing economic losses and reducing reliance on chemical interventions (Sanches and Wise 2020). Repeated use of substances acting in the same mode increases the risk of resistance, a common issue in pest management. Recent studies have

highlighted the role of endosymbionts in facilitating insecticide resistance (Zhang *et al.* 2022). Specifically, the gut microbiome of the BTM may contribute to resistance. The larvae's detoxification system, which protects them from plant-derived toxins, may also aid in neutralizing insecticides, thereby promoting resistance. Understanding these mechanisms is essential for developing strategies to inhibit them and improve the effectiveness of chemical control measures. This is a promising area of research for developing more effective pest control methods. Current studies on the box tree moth microbiota, including initial investigations in Bulgaria that identified five bacterial and fungal species (Sree and Varma 2015; Harizanova *et al.* 2018), underscore the need for further research. These studies hold significant potential for elucidating the microbiota's role in digestive processes and the detoxification of gut contents.

Chemical control remains a key component in managing box tree moths. However, the risk of resistance development underscores the necessity of implementing integrated pest management (IPM) strategies that combine chemical, biological, and mechanical approaches. A deeper understanding of detoxification mechanisms and the role of the gut microbiome in resistance is crucial for optimizing chemical control methods and ensuring their long-term effectiveness. Future research should prioritize the discovery of novel compounds with distinct modes of action and explore the potential of microbiome-targeted interventions to mitigate resistance.

Biological control and natural enemies

Biological control methods can serve as an effective complement to other long term strategies for managing the box tree moth. These methods help restore the natural balance of ecosystems and reduce the negative effects of chemical treatments. However, their effectiveness can be influenced by several factors, such as environmental conditions and the availability of natural enemies of the pest.

Biopesticides offer a more sustainable and environmentally friendly approach to pest control, although they may act more slowly and be less versatile than some chemical treatments. There are several non-chemical methods worth implementing to reduce the risk of box tree moth population resurgence. The lack of effective natural box tree moth enemies in invaded areas requires various pesticides for its control. Countries have been developing their control programs that prioritize biological preparations particularly those based on *Bacillus thuringiensis* var. *kurstaki* (Lacey *et al.* 2015) or baculoviruses (Rose *et al.* 2013). Biological agents containing *Bacillus thuringiensis* (Bt) strains are registered for controlling the moth's larvae.

The mechanism of action involves the bacteria producing crystalline protein inclusions during sporulation. These inclusions dissolve in the alkaline environment of the larvae's gut, releasing insecticidal toxins (Malinowski 2000). Bt-based products which are available for managing the box tree moth contain *B. thuringiensis* var. *kurstaki*, or *B. thuringiensis* var. *aizawai* and are found in granular or powdered forms, to be mixed into a water suspension. They act as midgut poisons to the moth larvae. After ingesting a treated leaf, the bacteria produce toxic proteins in the larvae's digestive system, which bind to receptors in the insect's gut, causing paralysis and eventually gut perforation, leading to death. The larvae stop feeding within 12-24 hours and typically die within 24-72 hours. In Poland among approved methods of biological control against the BTM, some products contain *B. thuringiensis* var. *kurstaki* (EG-2348, ABTS-1857, SA-11 and ABTS-351 strains) and others contain *B. thuringiensis* var. *aizawai* (ABTS-1857 strain). A significant advantage of these products is their selectivity – targeting only caterpillars of moths without harming beneficial insects like bees, ladybird bugs, or predatory insects. Moreover, these products are safe for humans, animals, and the environment, making them an ideal solution for sustainable agriculture. Using Bt reduces the reliance on conventional chemical pesticides, which benefits both the environment and human health. These products have demonstrated the highest effectiveness in controlling the early larval stages of the box tree moth, however, their efficacy can be affected by environmental conditions, foliage density, and the requirement for repeated applications throughout the growing season.

For controlling *C. perspectalis* entomopathogens are promising. These include fungi such as *Beauveria bassiana*, nematodes (e.g. *Steinernema carpocapsae* and *Heterorhabditis bacteriophora*), as well as baculoviruses (Göttig and Herz 2018; López *et al.* 2022; Miladinović *et al.* 2022; Yaman 2023). Entomopathogenic fungi involve spores adhering to the insect larvae's cuticle, germinating, and penetrating the insect's body. The fungus then proliferates inside the host, releasing toxins that eventually cause its death. *B. bassiana* products can be applied as sprays on plants, and thorough coverage of leaves is crucial to ensure contact between the spores and the larvae. Laboratory and field studies conducted by Iranian researchers have demonstrated that *B. bassiana* can achieve control rates of 66-100%, depending on concentration and exposure time (Zamani *et al.* 2023). In Poland, field trials have been conducted to test the effectiveness of biopreparations containing *Bacillus thuringiensis* var. *kurstaki* and *Beauveria bassiana* for controlling first- and second-generation larvae of *C. perspectalis*. The results indicated that the bacterial

formulation was more effective (90% mortality) than the entomopathogenic fungi, which caused 65% larval mortality (Bereś *et al.* 2020).

Additional research has explored the use of the fungus *Isaria fumosorosea*, although its efficacy is significantly lower than that of *B. bassiana* (Yaman 2023). Work with *Metarhizium anisopliae*, a pathogen effective against many insects, requires further investigation to assess its potential in controlling the box tree moth. Continued research and development of fungal-based biopesticides may contribute to more effective control of this invasive pest in the future.

As for baculoviruses, naturally occurring viruses have yet to be isolated from *C. perspectalis*. However, studies with other insect viruses from the genus *Alphabaculovirus* have demonstrated high larval mortality rates exceeding 90% (Rose *et al.* 2013; Oberemok *et al.* 2017; Yaman 2023).

In summary, biopesticides, particularly those using *Bacillus thuringiensis*, provide an effective, safe, and environmentally friendly solution for combating moth larvae. Their selectivity and low risk to non-target organisms and the environment make them an especially attractive tool for plant protection. Additionally, the use of biopesticides presents a lower risk of pests developing resistance due to their complex mechanisms of action. This complexity makes it harder for pests to adapt than to traditional chemical insecticides. Furthermore, the risk of phytotoxicity is lower than to some chemical products.

The box tree moth poses a serious threat to boxwood plants in Europe. Both in its native range and in Europe, no effective natural enemies have been identified to regulate this pest's populations in the field. However, various predators and parasitoids of the moth's larvae and eggs have been documented in regions of China, Japan, and Korea (Wan *et al.* 2014). Research has been ongoing for several years, mainly focusing on parasitic wasps, such as egg parasitoids of *Trichogramma* genus, (Göttig and Herz 2016; Can *et al.* 2022), as well as tachinid flies (*Tachinidae*) that parasitize the moth larvae (Wan *et al.* 2014; Martini *et al.* 2019). In Europe, particularly Switzerland, the presence of *Pseudoperichaeta nigrolineata* (*Tachinidae*) and *Apechthis compuctator* (*Ichneumonidae*) has been confirmed, indicating their potential for controlling this pest (Wan *et al.* 2014).

There have also been occasional reports of birds, such as tits, wagtails, blackbirds, starlings, and sparrows, attacking box tree moth larvae, though often rejecting them due to alkaloids present in their body, which likely deter predation (Leuthardt and Baur 2013; Mostini 2018). Among predatory birds, the *Ficedula albicollis* has been identified as a natural enemy, attacking the light moths, which, according to Bakay *et al.*

(2018) are more attractive to the bird than the darker forms (Bakay and Kollár 2018; Bereś *et al.* 2024). Instances of bird attacks on the moth have also been observed in Poland, though further research is needed to determine the role of birds in regulating moth populations.

Botanical insecticides

Recent studies have shown promising results regarding the use of natural plant-derived substances for pest control. Gokturk *et al.* (2021) investigated the insecticidal efficacy of essential oils from various plants, including *Origanum vulgare*, *O. onites*, *O. syriacum*, *O. majorana*, *Artemisia absinthium*, *Cuminum cyminum*, *Mentha pulegium*, and *Satureja hortensis*, against the larvae of *Cydalima perspectalis*. Their findings revealed that oregano essential oil demonstrated the highest effectiveness, resulting in larval mortality rates of 71-80%, while *Mentha pulegium* (field mint) was identified as the least toxic option. These studies suggest that essential oils may offer an effective and eco-friendly solution for controlling this pest (Gokturk *et al.* 2021). Additionally, essential oils could protect by preventing egg-laying and subsequent larval damage. Behavioral tests have shown that essential oils from cinnamon, eucalyptus, and lavender can repel *C. perspectalis* females from laying eggs, with cinnamon oil demonstrating the strongest repellent effect (Szelényi *et al.* 2020).

Conclusions and future perspectives

Cydalima perspectalis, commonly known as the box tree moth, represents a severe threat to boxwood cultivation and native ecosystems across Europe and beyond. The rapid spread of this invasive species and its significant impact on boxwood populations highlight the urgent need for effective and sustainable management strategies. Current methods should prioritize biological controls over chemical insecticides, particularly avoiding highly hazardous pesticides (HHPs) and broad-spectrum options like pyrethroids, which are both environmentally harmful and relatively ineffective against the BTM. To effectively target this pest, pheromone traps are recommended for precise timing of sprays with *Bacillus thuringiensis* formulations, while Spinosad offers an effective solution against mature larvae. Expanding research into the moth's natural enemies, enhancing environmentally friendly insecticides, and exploring the genetic diversity and larvae adaptability, including their interaction with microbial communities, could also provide valuable alternatives to conventional chemical controls.

As climate change and globalization contribute to the spread of the BTM, raising awareness among gardeners, landscape architects, and plant nursery professionals is essential for prompt identification and control. Monitoring efforts should extend beyond boxwood to other potential host plants, emphasizing a proactive approach to the moth's adaptability to new species. This shift toward integrated pest management, grounded in biological, mechanical, and minimally harmful chemical methods, is critical for preserving both cultivated and natural landscapes.

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