Insect pest management in hemp in Virginia

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Abstract

For the first time in many decades, a hemp pilot program was initiated in Virginia in 2016. Outdoor surveys were conducted in the 2017 and 2018 field seasons to record insect presence and feeding injury to plants. Multiple insect pests were present, including corn earworm (*Helicoverpa zea* [Boddie]) (Lepidoptera: Noctuidae), brown marmorated stink bug (*Halyomorpha halys* [Stål]) (Hemiptera: Pentatomidae), and cannabis aphid (*Phorodon cannabis*) (Hemiptera: Aphididae). In 2019, indoor production surveys revealed that cannabis aphid, twospotted spider mite (*Tetranychus urticae* Koch) (Acari: Tetranychidae), and hemp russet mite (*Aculops cannabicola* [Farkas]) (Acari: Eriophyidae) would likely cause production issues. Very little is known about the impact of insect defoliation in hemp so studies were conducted in 2018-2020 to determine impacts on yield and cannabinoid content of grain and cannabinoid variety hemp due to leaf surface area loss. In Virginia over two growing seasons, manual removal of leaf tissue in grain and CBD cultivars did not significantly impact observable effects on physical yield (seed or bud weight) or cannabinoid content (CBD or THC) at time of harvest. Corn earworm is the major pest of hemp produced outdoors and studies occurred to evaluate monitoring and management strategies. Pheromone traps may be valuable in determining when corn earworm moths are present in the vicinity of hemp fields but are not useful in predicting larval presence in buds or final crop damage.

Larval presence and final crop damage are related. Brown marmorated stink bug does not appear to be a concern in hemp, at least at this time.

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General audience abstract

For the first time in many decades, a hemp pilot program was initiated in Virginia in 2016. Outdoor surveys were conducted in the 2017 and 2018 field seasons to record insect presence and feeding injury to plants. Multiple insect pests were present, including corn earworm, brown marmorated stink bug, and cannabis aphid. In 2019, indoor production surveys revealed that cannabis aphid, twospotted spider mite, and hemp russet mite would likely cause production issues. Very little is known about the impact of leaf area loss due to insect feeding in hemp so studies were conducted in 2018-2020 to determine impacts on yield and cannabinoid content of grain and cannabinoid variety hemp due to leaf surface area loss. In Virginia over two growing seasons, manual removal of leaf tissue in grain and CBD cultivars did not significantly impact observable effects on physical yield (seed or bud weight) or cannabinoid content (CBD or THC) at time of harvest. Corn earworm is the major pest of hemp produced outdoors and studies occurred to evaluate monitoring and management strategies. Pheromone traps may be valuable in determining when corn earworm moths are present in the vicinity of hemp fields but are not useful in predicting larval presence in buds or final crop damage. Larval presence and final crop damage are related. Brown marmorated stink bug does not appear to be a concern in hemp, at least at this time.

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Introduction: Production and pest management of hemp, *Cannabis sativa* L., in the United States

Hemp, a low-delta-9-tetrahydrocannabinol (THC) variety of *Cannabis sativa* L., is an important plant that has been connected to humanity in the form of food, fiber, and medicine for many thousands of years (McPartland et al. 2019). By definition, hemp contains less than 0.3% delta-9-THC by dry weight and anything in excess of this threshold is considered federally illegal. Hemp production was prohibited in the United States for much of the 20th century and much of the germplasm and general production knowledge has since been lost; thus, it can be considered as a new crop in the U.S. With the passage of the 2014 Farm Bill (U.S. H.R. 2642 – 113th Congress [113-333]), U.S. Federal law allowed for the legal study of hemp as an industrial product crop. Even more recently, the passage of the 2018 Farm Bill (U.S. H.R. 2 –115th Congress [115-334]) removed hemp from the Federal list of controlled substances and declared it a crop distinct from marijuana (or a high-THC variety of *Cannabis sativa*) while also allowing for the development of organic industrial hemp certifications.

The market for hemp products in the U.S. is ever-changing and still becoming established (Mark and Snell 2019). Grain and fiber hemp contribute to the greatest number of industrial product uses (Fike 2016), but as of 2020, the U.S. does not yet have adequate infrastructure to process large quantities of harvested material (Malone and Gomez 2019). These industrial types of hemp require specialized equipment for planting and harvesting large acreages, but are less labor intensive throughout the growing season. Injury potential from pests is relatively low, but the profit per acre is also low (Mark and Snell 2019). Hemp for grain or fiber will likely have the greatest appeal to row crop or large acreage farmers. Hemp grown for cannabinoids has a narrower range of uses (Adesina et al. 2020), yet currently comprises the majority of acreage and holds the greatest profit potential (Schluttenhofer and Yuan 2017, Jelliffe et al. 2020). Cannabinoid hemp can be grown without specialized machinery, but is very labor intensive to harvest and dry and frequently requires storage space until time of sale. The marketable portion is the bud, or floral inflorescence, which can be harvested individually and sold as raw material or processed for cannabinoid extraction. In other cases, whole or remaining plant material can be processed as a unit, known as biomass.

Due to federal prohibition since the early 20th century, hemp production and pest management research is only now taking place and few to no recommendations exist for best growth practices or successful pest management. Some production information exists from other countries (Struik et al. 2000, Cosentino et al. 2012, Amaducci et al. 2015) and from the historic fiber production period in certain states (Wilsie et al. 1942, 1944, Ash 1948, Herndon 1963), but this provides little insight to the current situation in the U.S. as production is focused primarily on hemp for cannabinoids and acreage exists in almost every state. As most historical literature was focused on hemp for fiber production, statements were made that hemp rarely faced any pest issues (Boyce 1900, Herndon 1963), which is not the current case. As it stands in terms of production and pest management, some information on region-specific growing practices has been documented (Britt et al. 2020, Hansen et al. 2020), but much of the available literature is dated (Smith and Haney 1973, Miller 1982), does not focus exclusively on the U.S. (McPartland et al. 2000, McPartland and Rhode 2005, Punja et al. 2019), or profiles pests but does not make management recommendations (Lago and Stanford 1989, Cranshaw et al. 2018, 2019, Thiessen et al. 2020). The information shared from these resources has been helpful and profound in this early stage, but more targeted, recommendation-based information is certainly needed.

For the first time in many decades, an industrial hemp pilot program was initiated in Virginia in 2016. Outdoor surveys were conducted in the 2017 and 2018 field seasons to record insect presence and feeding injury to plants. Multiple insect pests were present on plants, including corn earworm (*Helicoverpa zea* [Boddie]) (Lepidoptera: Noctuidae), brown marmorated stink bug (*Halyomorpha halys* [Stål]) (Hemiptera: Pentatomidae), and cannabis aphid (*Phorodon cannabis*) (Hemiptera: Aphididae). In 2019, indoor production surveys revealed that cannabis aphid, twospotted spider mite (*Tetranuchys urticae* Koch) (Acari: Tetranychidae), and hemp russet mite (*Aculops cannabicola* [Farkas]) (Acari: Eriophyidae) would likely cause production issues. Not all species recorded in hemp are a concern (Table 1.1). Further information on specific arthropod pests in Virginia is available in Appendix I. A comprehensive review of arthropod pests of hemp and cannabis in the United States, including Virginia, was published by Cranshaw et al. in 2019.

In this dissertation, I report results from several studies that will help improve current knowledge and management of arthropod pests of hemp. Specific objectives include:

1. Assess impacts of corn earworm, *Helicoverpa zea*, on outdoor hemp and use information gathered to aid in the development of an integrated pest management program.
2. Evaluate the efficacy of biological and conventional insecticides on corn earworm, *Helicoverpa zea*, in a lab setting.

3. Evaluate the efficacy of biological and conventional insecticides on corn earworm, *Helicoverpa zea*, in a field setting.
4. Determine if hemp can serve as a suitable host plant, assess feeding effects on grain hemp, and evaluate the efficacy of insecticides for management of brown marmorated stink bug, *Halyomorpha halys*.
5. Assess the impact of manual defoliation on yield and cannabinoid content in grain and cannabinoid hemp cultivars.

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Chapter 1: Pest management needs and limitations for corn earworm (Lepidoptera: Noctuidae), an emergent key pest of hemp in the United States

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Abstract

Corn earworm, *Helicoverpa zea* (Boddie), has emerged as an injurious insect pest to hemp, *Cannabis sativa* L., a crop newly reintroduced to the United States. Hemp presents a potential opportunity for economic gain but can be challenging to produce and the market is developing and unstable. One of the most notable production challenges is managing multiple insect pests, including corn earworm. Corn earworm is particularly damaging when it feeds on flower buds produced in cannabinoid varieties, creating extensive bud tunneling and wounds that allow entry of pathogens that can aid development and presence of bud rot. Of moderate concern is damage to seeds in hemp cultivars grown for grain but little risk is posed to fiber varieties. Ability to research the crop has only recently been allowed as production was largely suspended following World War II and, as such, there has been limited opportunity to develop information for empirically based pest management recommendations. Further complicating development of IPM strategies are regulatory challenges associated with providing registration support to add hemp to pesticide labels, as it was not formally recognized as a crop by U.S. regulatory agencies until late 2019. Research needs and challenges to develop effective integrated pest management programs for corn earworm on hemp are reviewed.

Key words: corn earworm, hemp, management

Introduction

Hemp (*Cannabis sativa* L. containing a delta-9 tetrahydrocannabinol [THC] concentration of 0.3% or less on a dry weight basis) has seen a tremendous increase in cultivated acres in the United States, beginning with language in the 2014 U.S. Farm Bill (U.S. H.R. 2642 – 113th Congress [113-333]) that allowed hemp production pilot programs and accelerating dramatically following legalization for commercial production in the 2018 Farm Bill (U.S. H.R. 2 –115th Congress [115-334]). In this new era of U.S. hemp production, there are new opportunities for profit as well as production and pest management challenges. Among these are arthropod pests, and several have been identified as potentially limiting to the crop, including exotic specialists such as cannabis aphid, *Phorodon cannabis* Passerini (Hemiptera: Aphididae) (Cranshaw et al. 2018), Eurasian hemp borer, *Grapholita delineana* (Walker) (Lepidoptera: Tortricidae) (McPartland et al. 2000), and the hemp russet mite, *Aculops cannabicola* (Farkas) (Acari: Eriophyidae) (McPartland and Hillig 2003). Among native North American species, corn earworm, *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae), has been the most damaging insect pest of outdoor hemp in all locations in the U.S. (McPartland et al. 2000, Britt et al. 2020, Hansen et al. 2020).

Corn earworm larvae primarily feed on fruiting and reproductive structures of a wide variety of crops (Ditman and Cory 1931). Similarly, in hemp, corn earworm feeds little on foliage and concentrates feeding on flower buds and seeds and its importance as a pest is related to crop type and end use (i.e., grain, fiber, cannabinoids). Corn earworm injury is most significant in cannabinoid hemp as it is attracted to and feeds on the cola or bud structures. The marketable portion is the bud, or inflorescence, which can be harvested individually and sold as raw material or processed to extract cannabinoids that are later infused into food or personal care products or sold as tinctures (Schluttenhofer and Yuan 2017). In other cases, whole or remaining plant material can be processed as a unit, known as biomass, although cannabinoid levels are far lower in these parts of the plants. In cannabinoid cultivars, extensive bud tunneling/girdling occurs, frass contaminates harvested material, and feeding wounds predispose buds to rot, particularly in humid climates. In grain and fiber varieties, one main stalk with a seed head is produced at the apical point. In grain varieties, seeds are the harvested material. In fiber varieties, seeds are usually not harvested, but are present as reproductive structures (Amaducci et al. 2015). Seed heads, containing floral reproductive structures and seeds, are produced in monoecious varieties or female plants in dioecious varieties and are attractive to corn earworm moths, serving as an oviposition site. Corn earworm has been observed on occasion consuming entire seeds or damaging seeds to the point that they are no longer viable (Figure 1.1), but management importance in grain hemp is of little concern. Stalks are the harvested portion in fiber varieties and damage potential is low. Corn earworm has been observed feeding on stalks only in experiments where larvae were caged on seed heads (Figure 1.2); similar stalk boring behavior has been observed in tobacco (Allen and Lawson 1962).

Adult corn earworm oviposition and associated larval damage to hemp occurs during plant reproductive growth phases, which occur in late summer as day length begins to decline. At this time corn earworm populations may be high, either by migrants or from moths that locally emerged on earlier crops favorable to the species, notably corn. Hemp begins flower production when there are less than 14 hours of daylight; in the U.S., this occurs in late summer and typically coincides with the third annual generation of corn earworm; a fourth generation can also occur in the southeast and may cause additional damage. Hemp provides a valuable host at this point in the season as most spring-planted annual host crops are no longer flowering. Adult female moths lay eggs in floral inflorescences (Figure 1.3) and emerged larvae feed and injure bud material. Plant damage from larval feeding allows the entry of plant pathogens that cause buds to appear dark and rotted (Figure 1.4), colloquially referred to as "bud rot". Bud rot compromises the quality and, thus, marketability of hemp material. After harvest, remaining larval presence and associated frass can interfere with cannabinoid extraction. To preserve economic return with cannabinoid hemp, corn earworm must be managed to prevent flower loss, bud rot, and contamination.

There are challenges to corn earworm pest management in hemp. Here, we examine the situation posed by this damaging agricultural pest on an emerging high-value crop, discuss factors that contribute to pest management challenges, highlight current research, and emphasize an integrated pest management (IPM) approach to address the issue.

Corn earworm distribution, biology, and pest status

Corn earworm is widespread throughout the western hemisphere and has long been a key pest of agricultural crops such as cotton, *Gossypium hirsutum* L.; soybean, *Glycine max* (L.) Merr.; sorghum, *Sorghum bicolor* (L.); corn, *Zea mays* L.; tobacco, *Nicotiana tabacum* L.; tomato, *Solanum lycopersicum* L.; and various fruiting vegetables (Ditman and Cory 1931, Cohen et al. 1988, Kuhar et al. 2006, Swenson et al. 2013, Reisig and Reay-Jones 2015, Olmstead et al. 2016, Reay-Jones et al. 2016, Bibb et al. 2018, Reay-Jones 2019). Corn earworm has multiple generations per year depending on latitude (Reay-Jones 2019) and overwinters as a pupa in the soil in warmer regions of the U.S. Historically, corn earworm was reported as not able to overwinter north of 40^{o} north latitude (Phillips and Barber 1929, Ditman and Cory 1931), but that may no longer be the case due to climate change.

Female moths deposit eggs singly on above-ground plant parts, especially reproductive structures (such as corn silks) or upper surfaces of leaves of numerous host plants (Neunzig 1969). A single female can lay up to 1,500 eggs in her lifetime (Akkawi and Scott 1984, Fitt 1989). Eggs hatch in two to four days and the entire life cycle (egg, larva, pupa, and adult) can be completed in approximately 30 days (Butler and Scott 1976). The larval stage typically has six instars, but five is not uncommon and seven to eight have been reported (Capinera 1969). Larvae are almost always found concentrated among reproductive plant structures such as blossoms, buds, and fruits, producing direct damage to the harvested portion of the crop (Ditman and Cory 1931).

Corn earworm is considered by many to be the most economically-important agricultural pest in North America because it causes economic damage to multiple crops, is widely distributed, and is able to migrate to northern regions each year (Capinera 1969, Fitt 1989). Its impact on hemp crops in the U.S. has not been well studied because this crop has been legalized only recently for widespread production. However, the insect has been the most conspicuous and damaging pest of hemp grown outdoors since renewed crop production in 2015.

Corn earworm chemical management and sampling methods in agronomic crops

In addition to cultural control methods (e.g., planting date in sweet corn, row spacing in soybean), corn earworm is managed with several chemical insecticides. In corn and cotton, transgenic hybrids that express *Bacillus thuringiensis* (Bt) toxins are the standard control method (Edgerton et al. 2012, Reisig and Reay-Jones 2015, Reay-Jones et al. 2016). Recently, widespread resistance to several Bt proteins has been documented (Dively et al. 2016, Liu et al. 2017, Bilbo et al. 2018, Reisig and Kurtz 2018, Kaur et al. 2019, Dively et al. 2020) and foliar insecticide usage has increased. One in-plant toxin, Vip3Aa, still provides excellent control (Burkness et al. 2010, Yang et al. 2019, Dively et al. 2020).

In tobacco, corn earworm occurs in a lepidopteran complex with the tobacco budworm, *Chloridea virescens* (Fabricius), an insect of similar appearance and habit. Moths of both species lay eggs in tobacco buds, flowers, and seed heads and are, therefore, an economic concern prior to the removal of reproductive structures mid-season via a process called topping.

Insecticides used against these pests increase resistance risk in later season hosts (Abney et al. 2007). In recent years there have also been several reports of tobacco budworm from hemp.

In soybean, sweet corn, and other crops, damaging populations of corn earworm are controlled largely by use of synthetic insecticides of several modes of action, notably pyrethroids, diamides, and spinosyns (Kuhar et al. 2020, Taylor and Laub 2020, Kemble et al. 2021; Table 1.1). Corn earworm resistance to pyrethroids has become an issue in some U.S. populations (Stadelbacher et al. 1990, Brown et al. 1998, Hutchison et al. 2007, Jacobson et al. 2009), but the diversity of insecticides labeled for vegetable and field crops minimizes the risk of resistance and crop losses due to this pest (Kuhar et al. 2006, 2010, Swenson et al. 2013, Adams et al. 2016).

Corn earworm larvae are sampled in soybeans and cotton using sweep nets or beat sheets to determine the need for control measures. In cotton, vegetables, and tobacco, visual inspection of plants is often used to sample corn earworm and other pests (Bauske et al. 1998, Kuhar et al. 2006). The timing of insecticide applications can be based on the presence of eggs, larvae, or plant damage (Kuhar et al. 2006). Detection of adults can be done by use of blacklight traps but more commonly is done by use of various pheromone-baited traps (Hartstack et al. 1982, Hoffmann et al. 1991, Coop et al. 1992, Guerrero et al. 2014). Trap catch number, or presence/absence alone, can provide knowledge of when moths are actively flying and insecticide applications can be timed accordingly (Coop et al. 1992). In sweet corn, pheromone trap catch can be used to detect moth presence, reproductive activity, and crop damage potential (Chowdhury et al. 1987, Latheef et al. 1991, Olmstead et al. 2016); however, using trap catch thresholds to accurately predict ear damage can be challenging (Olmstead et al. 2016).

Challenges to corn earworm management in hemp

Developing effective pest management programs for corn earworm in hemp currently faces major challenges due to lack of knowledge and experience and lack of effective registered insecticides.

Lack of knowledge and experience. Unlike the aforementioned field crops and vegetables, which have over a century of field production and research to build upon, there has been essentially no targeted research on corn earworm in hemp. Hemp was historically grown in the U.S. for grain and fiber prior to a production ban in the early 20th century (Fike 2016, Malone and Gomez 2019, Mark and Snell 2019). Some production information exists from other countries (Struik et al. 2000, Cosentino et al. 2012, Amaducci et al. 2015) and from the historic fiber production period in a few states (Wilsie et al. 1942, 1944, Ash 1948), but this provides little insight to the current situation as corn earworm and other insect pests were considered minor (Boyce 1900, Herndon 1963). In this current era, hemp is being grown in the majority of states and production is currently focused primarily on hemp for cannabinoids, as this has been perceived to have the greatest profit potential per acre (Schluttenhofer and Yuan 2017, Jelliffe et al. 2020). Thus, a significant time and production gap renders most existing information obsolete. The current emphasis must focus on corn earworm damage to cannabinoid hemp.

Recent observations have established generally the type of plant injury to hemp from corn earworm and its potential to cause significant crop injury. However, many details are needed to help establish economic thresholds for the insect, including establishing what crop growth stages are attractive and induce egg laying, identifying effective sampling methods to predict incipient outbreaks, evaluating the range of resistance among cultivars, and potential effects of natural enemies. Pest management information compiled in the current era has either not focused on the United States (McPartland et al. 2000, McPartland and Rhode 2005), only described pest species without making management recommendations (Lago and Stanford 1989, Cranshaw et al. 2019, Punja et al. 2019, Thiessen et al. 2020), or remained state-specific (Britt et al. 2020, Hansen et al. 2020). Although corn earworm has been extensively studied on a great number of other crops, better information on its association with hemp is needed to develop more targeted, recommendation-based management strategies.

Lack of effective registered insecticides. Because of the peculiar legal history in the United States, hemp was not recognized as a crop site by U.S. regulatory agencies until December 2019. Since that time a few specific insecticide products have had label modifications that now include hemp. Essentially all pesticides that have received federal registration on hemp are food crop tolerance exempt and the overwhelming majority of insecticidal products are either microbial or botanically derived (Table 1.2).

Several characteristics are needed in an insecticide to manage corn earworm in hemp. Most importantly, it must be efficacious and capable of effecting a high level of control of larvae feeding on buds and developing seeds. Developing corn earworm larvae are largely exposed on the exterior of hemp unlike other crops where larvae tunnel into fruits or ears. This may facilitate potential success with contact products and insecticides requiring ingestion. Presently registered insecticides used for CEW management on several crops are reviewed in Table 1.1.

Second, insecticides must be compatible with pollinators. Flowering hemp produces pollen and can be highly attractive to a wide range of bees (O'Brien and Arathi 2019, Flicker et al. 2020), including honey bees, particularly in areas where there is a dearth of pollen available in late summer. As hemp flowering coincides with corn earworm egg laying, insecticides must be compatible with actively visiting pollinators.

Third, a significant market for hemp products is in organic production. Emphasis should be made to develop registrations that can effectively manage corn earworm in organic production systems.

Finally, there must be a realistic chance that a specific product can be registered. This will likely be very heavily affected by the type of hemp crop being produced. Hemp with end use in cannabinoid production will probably be most difficult to register insecticides on. This requires various solvent extractions of the crop and can be used by humans as a marketed product that may be ingested, inhaled, or topically applied. Extensive residue testing will likely be required at considerable expense. Less restricted would be hemp crops grown for grain, which may be able to be categorized within Crop Group 20 that includes various oilseeds, such as sunflower, canola, and cottonseed. Hemp crops grown strictly for fiber, without any consumption by humans or livestock, should have fewer restrictions and may most easily receive registrations.

Chemical control will be an ongoing challenge due to the historically complicated status of hemp at the federal level as well as disparate state approaches. Although hemp is now a federally legal crop in the U.S. as of 2018, agrichemical companies have been slow to register products. As of 2021, the only insecticides registered on hemp include several naturally-derived biopesticides and oils that are food crop tolerance exempt (EPA 2020; Table 1.2). Among these, a few have demonstrated activity on corn earworm (Britt and Kuhar 2020, Doughty et al. 2020, Britt et al. 2021 [in review]). However, complication arises as not all federally-registered products are registered in every state and each state has its own way of regulating pesticide use (Cranshaw et al. 2019). It will take an enormous investment to do residue studies required to register any insecticides that require food crop tolerance, particularly for cannabinoid hemp which is used to produce a part of the plant that then undergoes extraction of the cannabinoid by various processes, which would have to be included in residue trials, to produce a product variously used by humans through ingestion, inhalation, and topically. In the near future, it is unlikely that any registered insecticides with food crop tolerances will be allowed on hemp for cannabinoid production. It may be possible for grain crops, as they are similar to other oilseed crops (e.g., sunflower, canola), or a purely fiber crop, as it is not consumable.

Nucleopolyhedroviruses (NPVs) are important natural pathogens of many lepidopteran species that can be broadcast on crop fields. *Helicoverpa armigera nucleopolyhedrovirus* (HearNPV) is a heliothine specific virus in the *Baculoviridae* family (Inceoglu et al. 2001). A larva must ingest the NPV occlusion bodies to become infected (Inceoglu et al. 2001) and the virus replicates inside the insect while it continues to feed for 5 to 7 days until death. Infected larvae will display multiple symptoms such as a swollen body and movement to the top of the plant canopy (Figure 1.5), which can enhance spread of viral particles. The larvae will burst open and release the progeny virus upon death where it can spread through the environment by rain, wind, and other arthropods and persist to be ingested by subsequent generations of larvae (Black et al. 2019). Healthy larvae can become infected through cannibalism of infected larvae (Inceoglu et al. 2001). This epizootic effect, which can persist more than 21 days after application (Black et al. 2019), makes NPVs a potential tool for caterpillar management. Moreover, NPV insecticides are benign to mammals and other arthropods, making them ideal for use in IPM programs (Inceoglu et al. 2001). NPV products have shown promise in field efficacy trials on cotton in Mississippi (Little et al. 2017), sweet corn in California (Natwick 2013), and on hemp in Virginia (Doughty et al. 2020, Britt et al. 2021 [in review]). There are potential increases in efficacy if NPVs are combined with other biological products (Britt et al. 2021 [in review]).

Spray applications of products containing *Bacillus thuringiensis* var. *kurstaki* or *aizawai* are a frequently utilized chemical control tactic. Unfortunately, the widespread adoption of transgenic Bt corn and cotton expressing Cry1A toxins for management of corn earworm, coupled with the lack of non-Bt refuge compliance and sufficient high-dose of Bt toxin (Reisig 2017), has led to Bt-resistant corn earworm populations (Britt and Kuhar 2020, Doughty et al. 2020, Britt et al. 2021 [in review]). This greatly complicates corn earworm management in hemp as sprayable Bt products have largely been rendered ineffective due to Bt-resistant corn earworm populations that have cycled through transgenic Bt corn and cotton over multiple generations (Dively et al. 2016, Reisig and Kurtz 2018, Yang et al. 2019). Chambers et al. (1991) identified a unique protein from the Bt strain *aizawai*, which could enhance efficacy against

Cry1ab resistant insects and Bt aizawai-based insecticides have provided better control in field tests (Britt and Kuhar 2020, Britt et al. 2021 [in review]). Field control efficacy with any of the Bt products does not compare with that of some conventional and organic insecticide standards like chlorantraniliprole or spinosyn (Doughty et al. 2020, Britt et al. 2021 [in review]), but these products are not approved for use in hemp.

Other biological compounds and pathogens have shown little promise. Azadirachtins, derived from extracts of seed from the neem tree, *Azadirachta indica* (Meliaceae), have a wide range of insect growth and behavioral effects on insects (Schmutterer 1990), but do not provide adequate control of corn earworm in corn (Harding et al. 2020a, 2020b). A number of entomopathogenic microbial agents infect corn earworm. The use of the entomopathogenic fungus *Beauveria bassiana* (Bals.-Criv.) Vuill. has likewise demonstrated low efficacy (Doughty et al. 2020, Harding et al. 2020b). More research is needed on the potential of proteobacteria-derived insecticides such as heat-killed cells and fermentation solids of the bacteria *Burkholderia* spp. or *Chromobacterium subtsugae* strain PRAA4-1. These compounds are believed to work by contact and ingestion to disrupt insect exoskeletons and interfere with molting (Asolkar et al. 2013). No field trial data exist with these compounds but only moderate efficacy is seen in laboratory bioassays (Britt and Kuhar 2020).

Future research needs

Corn earworm management in hemp will continue to be a challenge until more data are available and viable sampling and control methods are found. Chemical management can be an effective short-term tool, but it is only one component of a sustainable integrated pest management (IPM) plan. Developing an integrated approach is the ultimate goal of many hemp entomology programs. Individual components that are needed, and currently lacking, in hemp IPM are set forth below.

1. ***Determine pest impacts of corn earworm on hemp.*** Economic thresholds currently cannot be developed because the market value for hemp is ill-defined and unstable (Mark and Snell 2019) and the costs of effective management tools are not known. In the near term, meaningful work is underway to determine the relationship of larval density to damage and marketable yield, which will contribute to future action thresholds for corn earworm in hemp.

   In 2020, several collaborators (KEB, TPK, HJB, MP, KAK, OSA, MSF, TAT, SZ, DO, HD, JJ) monitored corn earworm in commercial or university hemp fields in Virginia (n=7), North Carolina (n=4), Alabama (n=4), Maryland (n=2), and Delaware (n=1) ranging in size from 0.1 to 20.2 hectares (0.5 to 50 acres). Beginning at first appearance of buds, each field was sampled weekly by examining 30 random buds and recording the number of corn earworm larvae. An average peak of 7.2 ± 2.0 corn earworm larvae was found per 30 buds with the highest counts occurring in mid-September at all locations.

   In these same fields, we assessed corn earworm damage to buds using a scale from 0-3 where 0 was no damage; 1 was visual damage present but bud still marketable; 2 was damage to 50% or less of bud material rendering it unmarketable; 3 was damage to 50% or more of bud material rendering it unmarketable. Regression analysis indicated that corn earworm larval density in buds during the week of 31 August was a significant predictor of average damage rating, and that sample densities greater than 2.5 larvae per 30 buds would be predicted to result in unacceptable crop damage (rating >1; Figure 1.6).

Still, the economic value of hemp will need to stabilize to justify costs of implementing management tactics, but these data form the beginnings of development of an economic threshold.

2. ***Develop effective monitoring tools and methods*.** Visual plant inspections of buds for corn earworm larvae and concomitant bud damage are the best options for monitoring this pest. However, inspecting complex flower buds on hemp plants for early-stage corn earworm larvae can be challenging and time consuming. Monitoring adult moth activity with traps would provide a more efficient sampling tool. We explored the feasibility of this strategy in 2020 at the same locations at which bud injury was assessed in 2020. Mesh Scentry® Heliothis pheromone traps (Gempler's, Janesville, WI) with a Hercon Luretape corn earworm lure (Hercon Environmental, Emigsville, PA) attached to the center of a string hanging below the bottom opening of the trap were used for monitoring adult male corn earworm moths. Traps were mounted 1 meter above ground and were placed adjacent to hemp fields (within 1.5 to 3 meters from field edge) prior to hemp flowering. Lures were replaced every 14 days, and trap catch was recorded once per week from time of setup until harvest. Regression analyses were used to determine the relationship between weekly corn earworm moth catches and peak larval abundance on hemp. The total number of moths caught from one to four weeks prior to the peak larval abundance (mid-September) were used as predictors. Linear mixed-effects models were used with cumulative moth catch as a fixed effect and site nested within state as random effects. Larval abundance was

log10(x+1) transformed to improve data normality (Ives 2015). Analyses were performed using ‘Lmertest’ (Kuznetsova et al. 2017) package in R Statistical Software v3.5.2 (R Core Team 2018). No significant relationship was found between trap catch of moths and peak larval abundance for any of the four weeks that were assessed prior to the larval peak (Table 1.3). This suggests that pheromone trap catch of adult males may not be a reliable predictor of larval corn earworm presence in hemp. Additional research with different trap types of methodology may be worth exploring (Guerrero et al. 2014). In addition, understanding the impact of wind patterns, surrounding crops, and ovipositional preference of female moths may also provide some insight into the relationship between trap catch of male moths and subsequent larval damage in a specific crop (Wilson and Morton 1989, Latheef et al. 1991, Westbrook et al. 1995). As it stands, the best way to make an informed decision in regards to corn earworm pest management in hemp will be through visual inspection of flower buds.

3. ***Enhance use and conservation of natural biological control agents.*** Given the limited use of broad-spectrum insecticides in hemp, natural enemies have a high potential to impact pest populations. Cranshaw et al. (2019) and Schreiner (2019) have reported many natural enemy species in hemp, some of which are important natural enemies of corn earworm larvae and eggs on other crops such as *Coleomegilla maculata* (De Geer) (Coleoptera: Coccinellidae) (Seagraves and Yeargan 2009), *Orius insidiosus* (Say) (Hemiptera: Anthocoridae) (Peterson et al. 2018), and *Geocoris punctipes* (Say) (Hemiptera: Geocoridae) (Tillman and Mullinix 2003). It is important to understand the impact of these natural enemies in hemp to develop appropriate management recommendations. Furthermore, the

impact of other natural enemies such as egg parasitoids (*Trichogramma* spp. [Hymenoptera: Trichogrammatidae] and *Telenomus* spp. [Hymenoptera: Platygastridae]), larval parasitoids (*Archytas marmoratus* [Townsend] [Diptera: Tachinidae], *Toxoneuron nigriceps* Viereck [Hymenoptera: Braconidae], *Campoletis sonorensis* [Cameron] [Hymenoptera: Ichneumonidae]), and other generalist species has yet to be determined in hemp (Reay-Jones 2019). Conservation and inundative biological control efforts have increased natural enemy abundance and control of corn earworm in other crops such as sweet corn (Manandhar and Wright 2015, 2016).

4. ***Determine impacts of cultural control tactics.*** There is considerable phenotypic diversity among hemp types and varieties, but differences in pest susceptibility and host plant resistance have, to date, not been evaluated. Manipulation of planting and harvest dates should be evaluated as a way to minimize corn earworm exposure. Trap cropping with a crop of lower economic return is potentially valuable to deter pest presence from hemp, but complications arise with the instability of hemp economics. Corn earworm populations in hemp may be influenced by proximity to other crops, particularly flowering crops, but this has not yet been evaluated. Understanding spatial and temporal dynamics of this insect would be important to aiding greater understanding of risk factors for corn earworm infestation in hemp.

5. ***Determine efficacious insecticide options.*** Research is needed to explore the efficacy of existing options and optimize applications. Timing of applications should be tested in addition to combining or tank-mixing multiple insecticides. For instance, researchers have demonstrated enhanced efficacy of combining *Bacillus thuringiensis* with nuclear

polyhedrosis virus-based insecticides on *Helicoverpa armigera* (Huebner) (Lepidoptera: Noctuidae) (Liu et al. 2006, Marzban et al. 2009).

6. ***Encourage testing and registration of conventional lepidopteran insecticides on hemp.*** Diamide and spinosyn insecticides have become popular products for management in other crops including tobacco (Table 1.1), which, in many ways, holds the same niche crop use as hemp (i.e., it is a smokable product, harvested plants can be hung and dried, extractions of nicotine can be performed). These insecticides would provide critical control tools and can be IPM compatible because of their reduced impact on arthropod natural enemies (Chapman et al. 2009, Whalen et al. 2016).
7. ***Encourage government- and industry-partnered financial support to address challenges.*** Currently, very few states have positions at land-grant universities dedicated to hemp research and/or Extension activities. As a result, the demanding Extension responsibilities of a new and popular crop fall within the current duties of Extension specialists who cannot provide much of their time to hemp, particularly with minimal financial support. If crop acreage continues to increase and funding opportunities become available, land-grant universities should consider developing research, Extension, or joint positions to serve the needs of this new, rapidly growing industry. In this way, hemp-focused research and education can be shared with state-level officials, consultants, stakeholders, and producers.

Parting thoughts

Corn earworm will remain a damaging pest in hemp for the foreseeable future as currently there are a lack of effective monitoring and management tools. The best option for management at this time is to implement weekly scouting of larvae in flower buds and initiate control measures at first appearance of larvae. Among the limited insecticide options, nucleopolyhedrovirus insecticide products may offer the highest level of corn earworm larval suppression. Initiation of spray applications at first detection is critical to target early instars. Sprays are likely needed in weekly intervals until harvest to ensure that larvae encounter virus particles. As monitoring and management strategies are further evaluated in the coming years, successful and more sustainable management strategies will follow.

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Table 1.1. Selected insecticides registered in 2020 in the U.S. on host crops co-occurring with hemp for control of *H. zea.*

Active Ingredient (Group[1])	Product Name(s)[2]	Soybean	Tomato	Sweet corn	Tobacco	Cotton
Methomyl (1A)	Lannate	x	x	x	x	x
Alpha-cypermethrin (3A)	Fastac EC	x	x	x		x
Beta-cyfluthrin (3A)	Baythroid XL	x	x	x		x
Beta-cyfluthrin (3A) + Imidacloprid (4A)	Leverage 360	x	x			x
Bifenthrin (3A)	Brigade, Bifenture, Sniper, others	x	x	x	x	x
Bifenthrin (3A) + Imidacloprid	Brigadier, Swagger	x	x		x	x
Bifenthrin (3A) + Chlorantraniliprole (28)	Elevest	x	x	x		x
Cyfluthrin (3A)	Tombstone	x	x	x	x	x
Fenpropathrin (3A)	Danitol		x			x
Gamma-cyhalothrin (3A)	Proaxis,	x	x	x	x	x

[1] Insecticide group following Nauen et al. (2012): 1A = carbamate, 1B = organophosphate, 3A = pyrethroid, 4A = neonicotinoid, 5 = spinosyn, 6 = avermectin, 11 = midgut membrane disruptor, 18 = diacylhydrazine, 22 = oxadiazine, 28 = diamide.
[2] Product names mentioned are for convenience only. No endorsement of products is intended, nor is criticism of unnamed products implied.

Active Ingredient (Group[1])	Product Name(s)[2]	Soybean	Tomato	Sweet corn	Tobacco	Cotton
Lambda-cyhalothrin (3A)	Warrior II, Karate LambdaCy, Lambda T, Silencer, or others	x	x	x	x	x
Lambda-cyhalothrin (3A) + Chlorantraniliprole (28)	Besiege	x	x	x	x	x
Lambda-cyhalothrin (3A + Thiamethoxam (4A)	Endigo	x	x		x	x
Permethrin (3A)	Permethrin 3.2EC, Perm-Up, or others		x	x		
Zeta-cypermethrin (3A)	MustangMax		x	x		x
Zeta-cypermethrin (3A) + Bifenthrin (3A)	Hero EC	x	x	x		x
Zeta-cypermethrin (3A) + Abamectin (6)	Gladiator		x			x
Spinetoram (5)	Radiant	x	x	x		x

Active Ingredient (Group[1])	Product Name(s)[2]	Soybean	Tomato	Sweet corn	Tobacco	Cotton
Spinetoram (5) + Methoxyfenozide (18)	Intrepid Edge	x	x[3]	x[4]		x
Novaluron (15)	Rimon	x	x	x		x
Spinosad (5)	Blackhawk, Entrust	x	x	x	x	x
Emamectin benzoate (6)	Proclaim		x			
Bacillus thuringiensis kurstaki (11)	Dipel, Javelin, others	x	x	x	x	x
Bacillus thuringiensis aizawai (11)	Agree, Xentari	x	x	x	x	x
Methoxyfenozide (18)	Intrepid		x			x
Indoxacarb (22)	Steward, Avaunt[5]	x	x	x		x
Chlorantraniliprole (28)	Prevathon, Coragen, Vantacor	x	x	x	x	x
Cyantraniliprole (28)	Exirel, Verimark	x[6]	x	x	x	x
Cyclaniliprole (28)	Harvanta		x			
Cyantraniliprole (28) + Abamectin (6)	Minecto Pro		x			x

[3] Only registered for use in CA
[4] Not registered for use in NY, and not permissible in CA & AZ
[5] Can only be applied in sweet corn prior to silking
[6] Foliar (Exirel – FMC allows, Dupont does not allow), Furrow (Verimark does not allow)

Active Ingredient (Group[1])	Product Name(s)[2]	Soybean	Tomato	Sweet corn	Tobacco	Cotton
Polyhedral occlusion bodies of *Helicoverpa zea* nucleopolyhedrovirus	Heligen, Gemstar, Helicovex	x	x	x	x	x

Table 1.2. Pesticide active ingredients used to control arthropod pests that have one or more formulations registered by the United States Environmental Protection Agency that include hemp on the label (as of 1 February 2021).

Active Ingredient[7]	**No. products registered**	**Potential value in CEW management**
Autographa californica Multiple Nucleopolyhedrovirus strain R3	1	None
Azadirachtin	3	Low
Azadirachtin + Neem oil	3	Low
Bacillus thuringiensis subsp. *aizawai* strain GC-91	1	Moderate
Bacillus thuringiensis subsp. *kurstaki* strain -03	1	Moderate
Bacillus thuringiensis subspecies *kurstaki*, strain EVB	1	Moderate
Beauveria bassiana strain ANT-03	2	Low
Beauveria bassiana strain GHA	2	Low
Chromobacterium subtsugae strain PRAA4-1 cells and spent fermentation media	1	None
Chrysodeixis includens Nucleopolyhedrovirus isolate #460	1	None
Chrysodeixis includens Nucleopolyhedrovirus isolate #460 & Helicoverpa zea (including mixtures with additional viruses)	3	Low

[7] Additional commercial products containing more than one of the listed active ingredients are also registered.

Active Ingredient[7]	No. products registered	Potential value in CEW management
GS-omega/kappa - Hxtx-Hv1a (spider venom peptides)	1	Unknown
Heat-Killed *Burkholderia* sp strain A396 cells and spent fermentation media	1	None
Helicoverpa armigera Nucleopolyhedrovirus strain ABA-NPV-U	1	Moderate-High
Isaria fumosorosea Apopka strain 97	1	None
Neem oil	3	None
Polyhedral occlusion bodies of *Helicoverpa zea* Nucleopolyhedrovirus ABA-NPV-U	1	Moderate-High
Polyhedral occlusion bodies of the nuclear polyhedrosis virus of *Helicoverpa zea* (corn earworm)	1	Moderate-High

Table 1.3: Predicting larval abundance of corn earworm in early September using accumulated moth counts from various trapping periods from first moth detection. (Log-transformed mixed effects)

Final Week	**Term**	**Estimate ± s.e**	**Df**	**t-value**	**p-value**	**r^2**
Aug. 17	Intercept	1.83 ± 0.45	3.7	4.09	0.02*	0.83
	# moths	0.003 ± 0.005	7.8	0.58	0.58	
Aug. 24	Intercept	1.86 ± 0.38	10.1	4.92	0.0001*	0.82
	# moths	0.001 ± 0.001	9.2	0.84	0.42	
Aug. 31	Intercept	1.65 ± 0.38	5.3	4.35	0.006*	0.64
	# moths	0.001 ± 0.001	9.5	1.06	0.31	
Sept. 7	Intercept	1.67 ± 0.38	5.3	4.44	0.005*	0.63
	# moths	0.001 ± 0.001	6.6	0.97	0.36	

Figure 1.1: Hemp seed devoured by corn earworm.

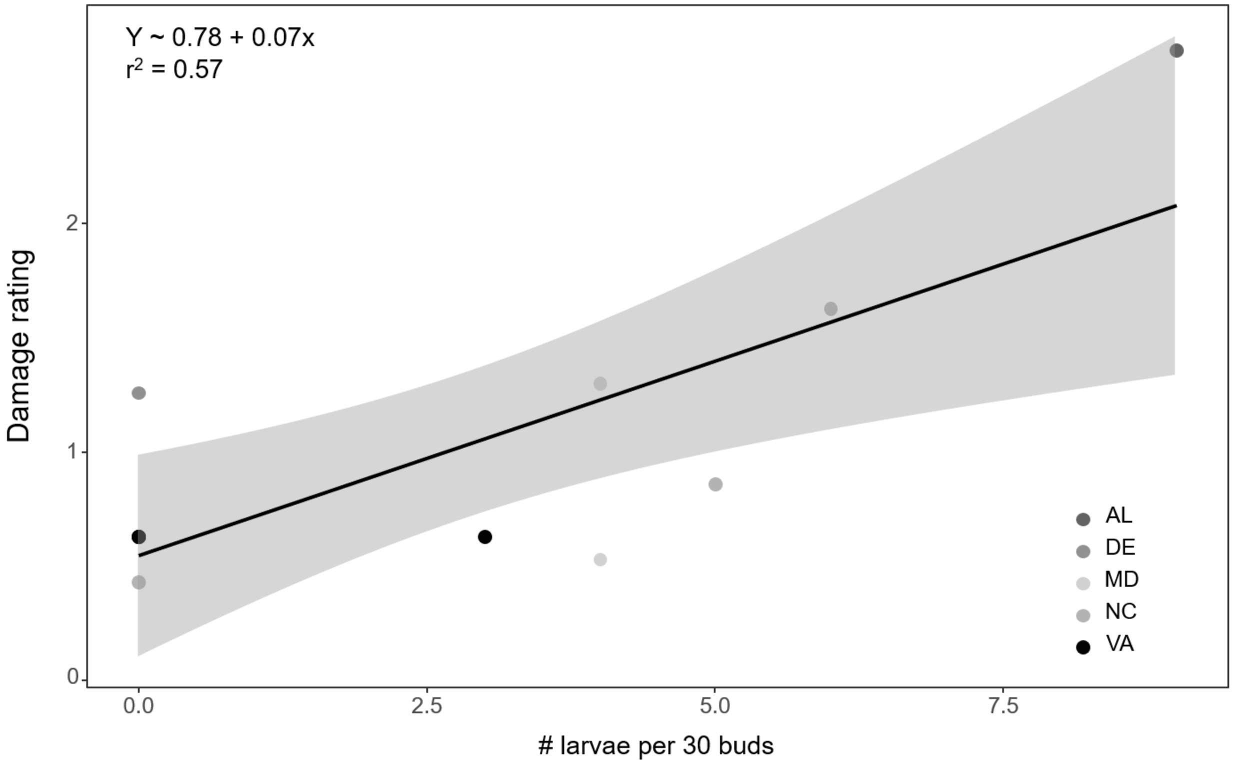
Figure 1.2: Corn earworm larva tunneled into hemp stem.

Figure 1.3: Corn earworm eggs laid in hemp bud.

Figure 1.4: Bud rot in hemp with corn earworm present.

Figure 1.5: Corn earworm larva infected with *Helicoverpa zea* nucleopolyhedrovirus insecticide.

Figure 1.6: Regression analysis of damage rating and peak larval presence in hemp buds.



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Chapter 2: Laboratory bioassays of biological/organic insecticides to control corn earworm on hemp in Virginia, 2019

(As published in Arthropod Management Tests: Britt, K. E., and T. P. Kuhar. **2020**. Laboratory bioassays of biological/organic insecticides to control corn earworm on hemp in Virginia, 2019. Arthropod Manag. Tests. 45: 1–2.)

Two separate bioassays were conducted in fall 2019 to evaluate the effects of biological/organic insecticide products on CEW in hemp. Bioassay 1 was initiated on 16 Sep 2019 and included the following treatments: Gemstar (*Helicoverpa zea* nuclear polyhedrosis virus [HzNPV]), Javelin (*Bacillus thuringiensis* var. *kurstaki*), DiPel (*Bacillus thuringiensis* var. *kurstaki*), XenTari (*Bacillus thuringiensis* var. *aizawai* + *kurstaki*), Venerate (94.5% Heat-killed *Burkholderia* spp. strain A396 cells and spent fermentation media), Grandevo (30% *Chromobacterium subtsugae* strain PRAA4-11 and spent fermentation media), Entrust (Spinosad), and an untreated check (Table 2.1). Third and fourth instar CEW larvae were collected from ears from an untreated field of sweet corn (*Zea mays*) established at Virginia Tech's Kentland Farm in Whitethorne, VA (Kentland). Only vigorous larvae with fresh color were used for the experiment. On 16 Sep 2019, hemp seed heads ('Felina-32') were collected from field plots at Kentland, brought to the laboratory, and cut into ~9 cm^3 sections. Forty hemp seed head sections were dipped into spray-tank concentrations of each treatment (Table 2.1) and placed individually into 1 oz plastic diet cups with a single CEW larva. A tray of 10 cups represented a replicate and four replicates were established for each treatment and placed in a different stack on the laboratory bench for the duration of the experiment. Diet cups were placed on the laboratory benchtop and held at laboratory ambient light and temperature (20–25°C) for 96 h and checked daily for mortality. Percent mortality data were analyzed with ANOVA procedures and means separated with Tukey's HSD.

Bioassay 2 was initiated on 2 Oct 2019 and included the following treatments: Agree (*Bacillus thuringiensis* var. *aizawai*), Javelin (*Bacillus thuringiensis* var. *kurstaki*), Deliver (*Bacillus thuringiensis* var. *kurstaki*), XenTari (*Bacillus thuringiensis* var. *aizawai* + *kurstaki*), PyGanic (pyrethrins), Entrust (spinosad), and an untreated check (Table 2.2). The experiment was conducted using the same aforementioned procedures except that rather than using field-collected CEW, which were depleted from the field, we used third instars raised on artificial diet that were purchased from Benzon Research Inc., Carlisle, PA.

In bioassay 1, CEW mortality varied greatly among treatments. As control mortality remained low (<15%) for the duration of the experiment, the 4 DAT data are probably the most useful because many of the treatments tested take a few days to actually kill larvae (Table 2.1, Figure 2.1). Entrust resulted in a significantly higher mortality than any other product with 95% mortality after 4 d. XenTari (67.5%) had a significantly higher mortality than any of the other *Bacillus thuringiensis* products and its efficacy was similar to Venerate (55%). Javelin and DiPel resulted in a significantly higher mortality (32.5% and 35%, respectively) than Gemstar (10%) and the untreated check (15%). It should be noted that Gemstar frequently takes more than 96 h to have a lethal effect on larger CEW larvae.

In bioassay 2, PyGanic and Entrust performed significantly better than all other treatments, resulting in 100% and 97.5% mortality, respectively (Table 2.2, Figure 2.2). XenTari, again, had the highest mortality among the tested *Bacillus thuringiensis* products (75%).

However, it only had significantly higher mortality than Agree (47.5%) and the untreated check (2.5%). Javelin and Deliver obtained 67.5% and 60% mortality, respectively, which was almost double what these treatments achieved with field-collected larvae in Bioassay 1. Resistance to Cry1AB Bt proteins is widespread in Virginia CEW populations, and likely explains this difference. It should be noted that resistance to pyrethroid/pyrethrin insecticides is also observed widely in Virginia CEW populations, and thus, although not tested in Bioassay 1, PyGanic would not be expected to result in 100% mortality of field-collected CEW larvae.

Table 2.1. Mean percentage mortality of field-collected corn earworm 3rd to 5th instars placed on hemp seed heads dipped in field-rate concentrations of various insecticide treatments in Blacksburg, VA, Virginia in 2019.

			Average % mortality			
Treatment	**Rate/acre**	**Rate/1500 mL water**	**1 DAT**	**2 DAT**	**3 DAT**	**4 DAT**
Untreated control			0.0 c	12.5 cd	12.5 de	15.0 d
Gemstar	5 fl. oz.	1.7 mL	2.5 c	2.5 d	5.0 e	10.0 d
Javelin	16 oz.	5.21 g	0.0 c	12.5 cd	25.0 cd	32.5 c
DiPel	16 oz.	5.21 g	5.0 c	17.5 cd	32.5 c	35.0 c
XenTari	16 oz.	5.21 g	12.5 abc	52.5 ab	57.5 b	67.5 b
Venerate	128 fl. oz.	43.5 mL	15.0 abc	30.0 bc	37.5 c	55.0 b
Grandevo	48 oz.	15.6 g	20.0 ab	22.5 cd	22.5 cde	22.5 cd
Entrust	5 fl. oz.	1.7 mL	27.5 a	65.0 a	87.5 a	95.0 a
P-value			0.0142	0.0004	0.0001	0.0001

Figure 2.1. Mean percentage mortality of field-collected corn earworm 3rd to 5th instars placed on hemp seed heads dipped in field-rate concentrations of various insecticide treatments in Blacksburg, VA, Virginia in 2019.

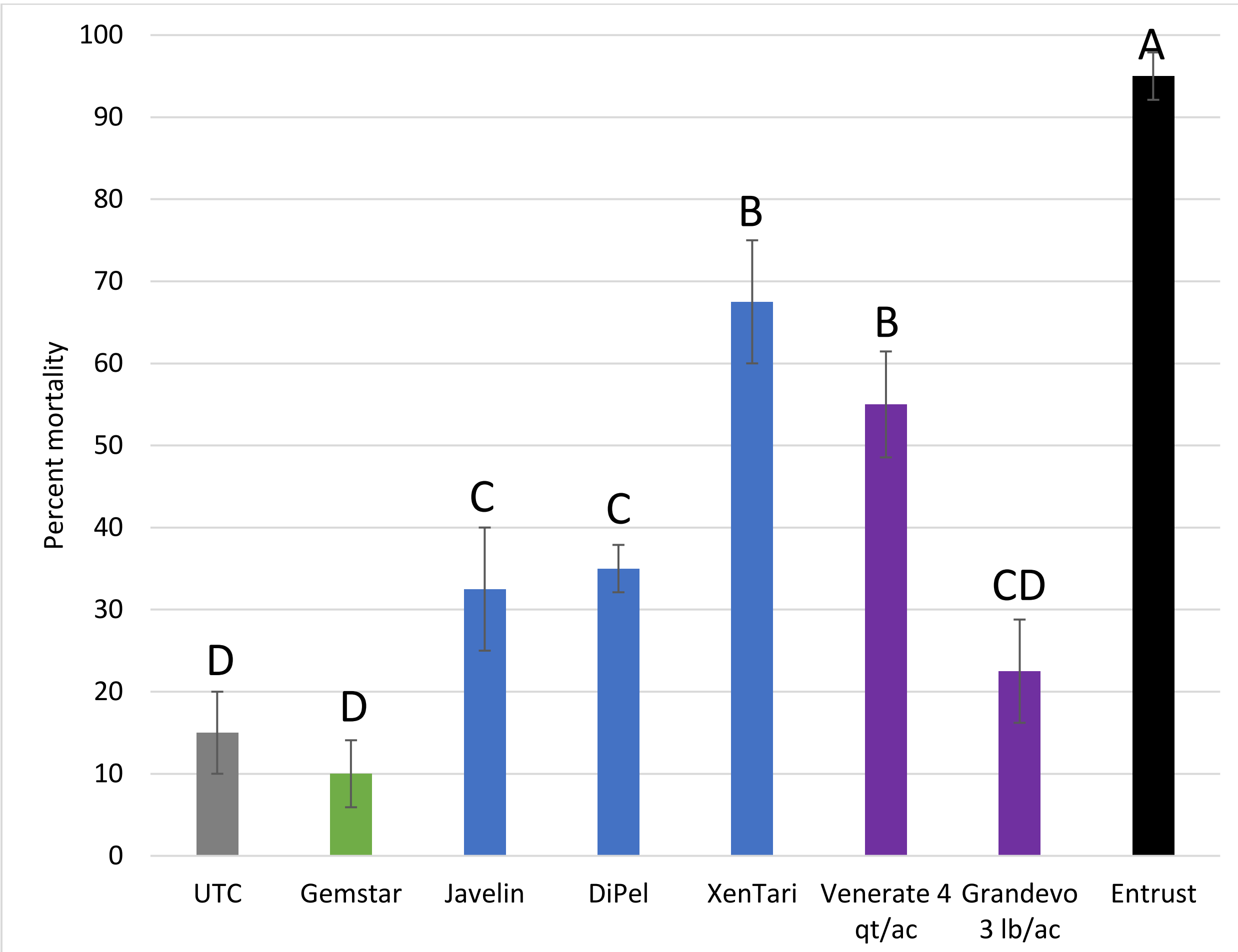
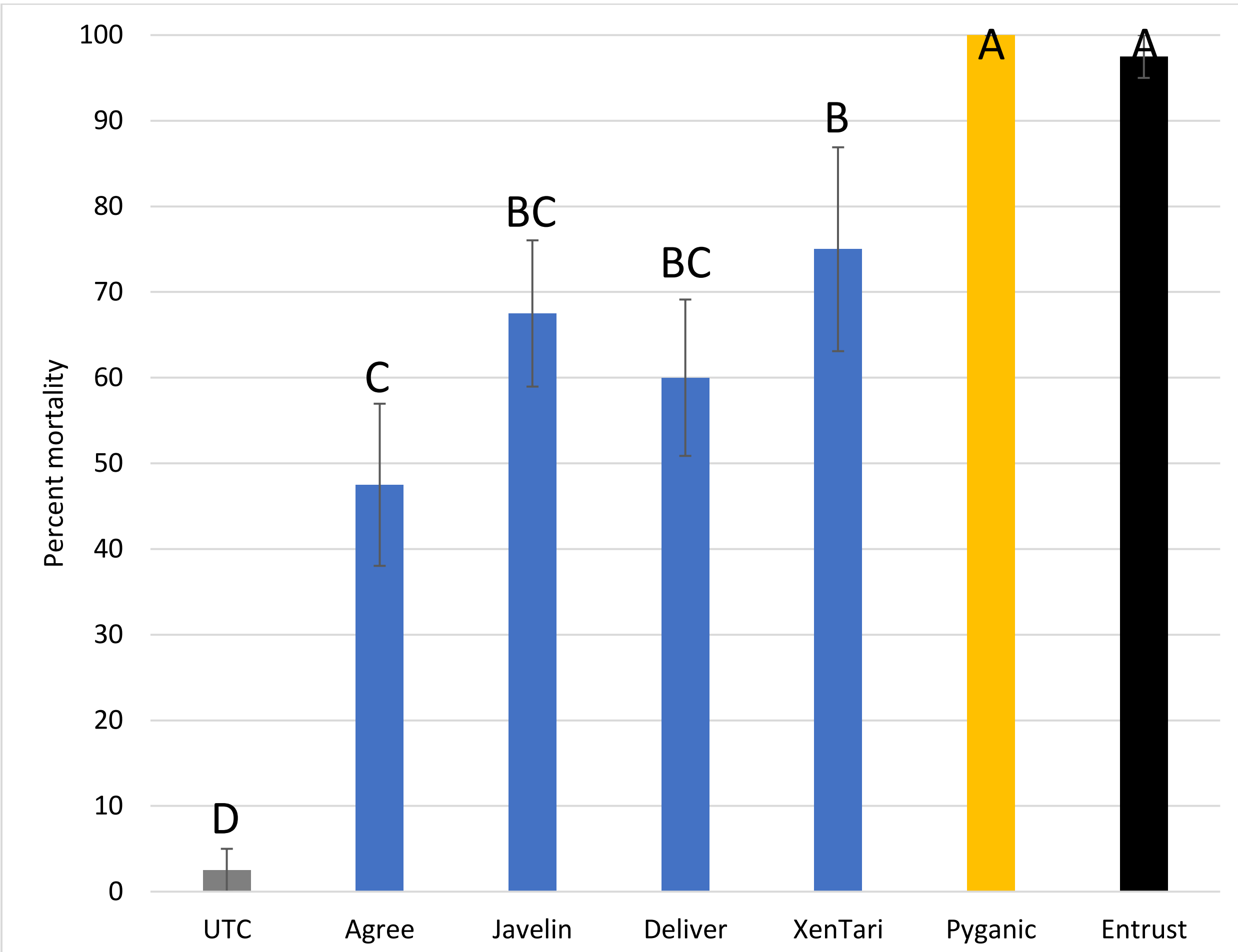


Table 2.2. Mean percentage mortality of laboratory-reared susceptible corn earworm 2nd to 3rd instars placed on hemp seed heads dipped in field-rate concentrations of various insecticide treatments in Blacksburg, VA, Virginia in 2019.

			Average % mortality			
Treatment	**Rate/acre**	**Rate/1500 mL water**	**1 DAT**	**2 DAT**	**3 DAT**	**4 DAT**
Untreated control			0.0 c	2.5 c	2.5 d	2.5 d
Agree	16 oz.	5.21 g	2.5 c	7.5 c	37.5 c	47.5 c
Javelin	16 oz.	5.21 g	0.0 c	10.0 c	55.0 bc	67.5 bc
Deliver	16 oz.	5.21 g	5.0 c	10.0 c	47.5 bc	60.0 bc
XenTari	16 oz.	5.21 g	5.0 c	12.5 c	62.5 bc	75.0 b
Pyganic	59 fl. oz.	20.0 mL	97.5 a	97.5 a	100 a	100 a
Entrust	5 fl. oz.	1.7 mL	37.5 b	82.5 b	92.5 a	97.5 a
P-value			0.0001	0.0001	0.0001	0.0001

Figure 2.2. Mean percentage mortality of laboratory-reared susceptible corn earworm 2nd to 3rd instars placed on hemp seed heads dipped in field-rate concentrations of various insecticide treatments in Blacksburg, VA, Virginia in 2019.



Chapter 3: Evaluation of biological insecticides to manage corn earworm in CBD hemp, 2020

(as submitted to Arthropod Management Tests: Britt, K. E., T. D. Reed, and T. P. Kuhar. **2021**. Evaluation of biological insecticides to manage corn earworm in CBD hemp, 2020. Arthropod Manag. Tests. 46.)

A field experiment was conducted with 'Sweeten' hemp transplanted into raised soil beds on 2 July 2020 at the Virginia Tech Southern Piedmont Agricultural Research and Education Center in Blackstone, VA. The experiment had seventeen treatments: Agree WG (*Bacillus thuringiensis* var. *aizawai*), Crymax (*Bacillus thuringiensis* var. *kurstaki*), Entrust SC (spinosad), Gemstar LC (Polyhedral occlusion bodies of the nuclear polyhedrosis virus of *Helicoverpa zea*), Gemstar LC + BoteGHA ES (Polyhedral occlusion bodies of the nuclear polyhedrosis virus of *Helicoverpa zea* + *Beauveria bassiana* strain GHA), Spear-Lep (GS-omega/kappa-Hxtx-Hv1a), Spear-Lep + Leprotec (GS-omega/kappa-Hxtx-Hv1a + *Bacillus thuringiensis* var. *kurstaki*), Heligen A (Polyhedral occlusion bodies of *Helicoverpa zea*; applied on 2 September 2020 ONLY), Heligen AB (Polyhedral occlusion bodies of *Helicoverpa zea*; applied on 2 and 8 September 2020 ONLY), Heligen ABC (Polyhedral occlusion bodies of *Helicoverpa zea* applied on 2, 8, and 15 September 2020), XenTari DF (*Bacillus thuringiensis* var. *aizawai*), Heligen + XenTari DF (Polyhedral occlusion bodies of *Helicoverpa zea* + *Bacillus thuringiensis* var. *aizawai*), PyGanic EC (pyrethrins), PyGanic EC + Exponent (pyrethrins + piperonyl butoxide), DiPel DF (*Bacillus thuringiensis* var. *kurstaki*), Coragen (Chlorantraniliprole), and an untreated check arranged in an RCB design with four replicates. Individual plots were comprised of 5 plants. Approximately one week after flowering, hemp plants were sprayed with insecticides in the field using a 3-nozzle boom equipped with D3 spray tips powered by a CO_2 back sprayer at 40 PSI. All treatments were applied 3 times: 2, 8, and 15 September 2020. On 8, 15, and 23 September, the number of CEW, virus-infected CEW, and presence of bud rot was counted on 10 buds per plot (Table 3.1). Data were analyzed using ANOVA procedures and means were separated using Tukey's HSD at the 0.05 level of significance.

On 8 September (7 DAT1), there was no treatment effect on number of CEW per 10 buds per plot (Table 3.1, Figure 3.1). On 15 September (7 DAT2), Gemstar LC + BoteGHA ES, Heligen ABC, and Coragen treatments had significantly fewer CEW than PyGanic EC and PyGanic EC + Exponent. Gemstar LC had significantly fewer CEW than PyGanic EC + Exponent. On 23 September (8 DAT3), Entrust SC had significantly fewer CEW than Crymax and the untreated check. PyGanic EC and PyGanic EC + Exponent likely negatively impacted natural enemies present in plots, thus allowing CEW numbers to increase higher than if plots were untreated. Both treatments had significantly more cumulative CEW than Entrust SC, Gemstar LC + BoteGHA ES, Heligen ABC, and Coragen. PyGanic EC + Exponent had significantly more cumulative CEW than Heligen + XenTari DF. Gemstar + BoteGHA resulted in the lowest number of cumulative CEW per plot; this treatment was not significantly different from Entrust SC, Heligen ABC, or Coragen. All treatments with NPV active ingredient had the greatest number of virus-infected CEW. Gemstar LC, Heligen A, Heligen AB, and Heligen ABC had significantly more virus-infected CEW than the untreated check; Gemstar LC + BoteGHA ES was not significantly different from the untreated check. Gemstar LC had a significantly higher presence of bud rot than Entrust SC, Gemstar LC + BoteGHA ES, Heligen A, and Coragen. Entrust SC had a significantly lower presence of bud rot than all treatments except the untreated check, Gemstar LC + BoteGHA ES, Spear-Lep, Heligen A, Heligen AB, and Coragen. No signs of phytotoxicity were observed from any treatments.

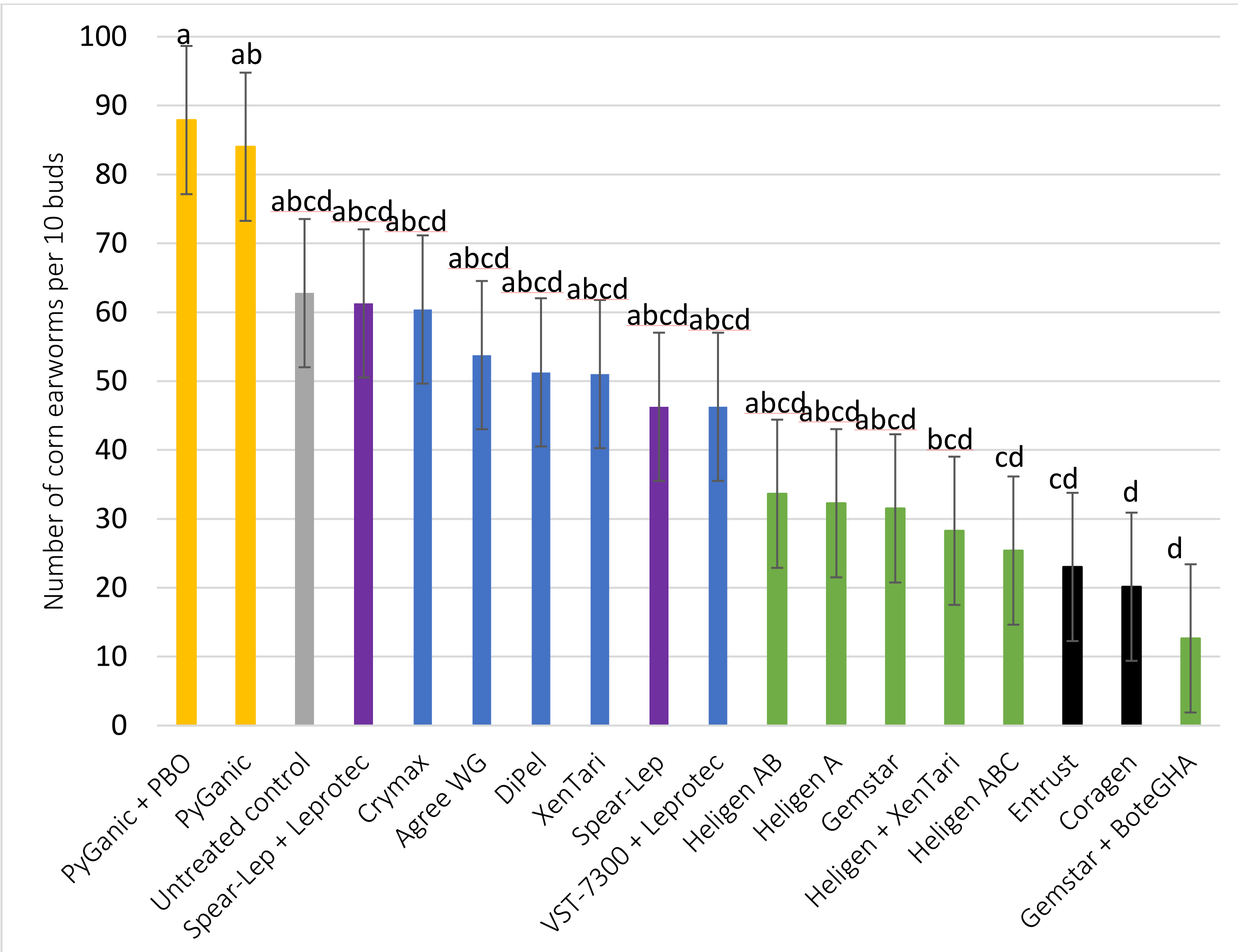
Table 3.1. Corn earworm larval densities, numbers of virus-infected larvae, and proportion of buds with rot from CBD oil hemp sprayed with various insecticide treatments in Blackstone, Virginia in 2020.

Treatment	Rate/acre	Number of CEW larvae per 10 buds			Cumulative CEW days	Cumulative virus-infected corn earworm per 10 buds	Proportion of buds with rot on 23 September 2020*
		8 September	15 September	23 September			
Untreated control	--	2.3 a	3.5 abc	3.8 a	62.9 abc	1.9 c	0.4 abcde
Agree WG	16.0 oz	1.5 a	3.8 abc	2.3 ab	53.8 abc		0.6 abc
Crymax	16.0 oz	1.8 a	3.8 abc	3.5 a	60.4 abc		0.5 abcd
Entrust SC	5.0 fl. oz	0.8 a	1.5 abc	0.0 b	23.0 c		0.1 e
Gemstar LC	5.0 fl. oz	1.3 a	1.0 bc	2.3 ab	31.5 abc	48.3 ab	0.8 a
Gemstar LC + BoteGHA ES	5.0 fl. oz 16.0 fl. oz	0.0 a	0.5 c	0.5 ab	12.7 c	33.5 bc	0.2 cde

Spear-Lep	32.0 fl. oz	2.3 a	2.5 abc	1.5 ab	46.3 abc		0.5 abcde
Spear-Lep + Leprotec	32.0 fl. oz 16.0 fl. oz	3.3 a	3.5 abc	1.8 ab	61.3 abc		0.6 abc
Heligen A	2.4 fl. oz	1.0 a	2.3 abc	0.5 ab	32.3 abc	63.0 ab	0.3 bcde
Heligen AB	2.4 fl. oz	1.0 a	1.5 abc	2.3 ab	33.7 abc	73.6 a	0.4 abcde
Heligen ABC	2.4 fl. oz	1.5 a	0.5 c	1.3 ab	25.4 c	52.8 ab	0.6 abc
XenTari DF	16.0 oz	2.3 a	3.0 abc	1.8 ab	51.0 abc		0.5 abcd
Heligen + XenTari DF	2.4 fl. oz 16.0 oz	1.3 a	1.5 abc	0.5 ab	28.3 bc	32.0 bc	0.5 abcd
PyGanic EC	15.6 fl. oz	3.3 a	6.0 ab	2.8 ab	84.0 ab		0.5 abcd
PyGanic EC + Exponent	15.6 fl. oz 23.0 fl. oz	3.3 a	6.3 a	3.3 ab	87.9 a		0.6 ab
DiPel DF	16.0 oz	2.3 a	2.5 abc	2.8 ab	51.3 abc		0.5 abcd
Coragen	3.5 fl. oz	1.0 a	0.5 c	0.8 ab	20.2 c		0.1 de

P-value from ANOVA	0.0233 (NS)	0.0015	0.0025	0.0001	0.0014	<.0001

Figure 3.1. Corn earworm larval densities from CBD hemp sprayed with various insecticide treatments in Blackstone, Virginia in 2020.



Chapter 4: Brown marmorated stink bug (Hemiptera: Pentatomidae) associated with *Cannabis sativa* (Rosales: Cannabaceae) in the United States and evaluation of insecticides to control it

(As published in the following two journal articles: Britt, K. E., M. K. Pagani, and T. P. Kuhar. 2019. First report of brown marmorated stink bug [Hemiptera: Pentatomidae] associated with *Cannabis sativa* [Rosales: Cannabaceae] in the United States. J. Integr. Pest Manag. 10: 1-3;

Britt, K. E., and T. P. Kuhar. 2020. Evaluation of insecticides to control brown marmorated stink bug on hemp in Virginia, 2019. Arthropod Manag. Tests. 45: 1-1.)

Abstract

Brown marmorated stink bug, *Halyomorpha halys* (Stål), is a highly polyphagous pest in North America and Europe. Herein, we report our observations of this invasive stink bug on grain hemp (*Cannabis sativa*) in Virginia, which, to our knowledge, is the first published report of *H. halys* associated with that crop. Effects of damage to hemp plants from this insect are unknown, so studies were initiated in 2018 to investigate further. Bugs were caged in varying densities for several weeks on seed heads of grain variety industrial hemp in field plots to document damage appearance and yield effects. Seeds were removed from plants in the laboratory, counted, and weighed to assess differences between treatments. In another study, bugs were reared on hemp seed heads in a lab setting from the second instar stage to adulthood. We found that bugs developed successfully to adulthood. Although further studies

are needed, it appears that at this time, *H. halys* may not be a threat to yield and quality of industrial hemp.

Introduction

The brown marmorated stink bug, *Halyomorpha halys* (Stål), is an invasive species from east Asia (Lee et al. 2013) that likely entered the United States in the mid-1990s, first detected in eastern Pennsylvania (Hoebeke and Carter 2003). Since the early 2000s, *H. halys* has spread throughout much of the United States, has established in Canada and several European countries, and has become a significant agricultural pest (Haye et al. 2015, Leskey and Nielsen 2018). *Halyomorpha halys* is a highly polyphagous pest with a broad host range of over 170 plant species including a wide array of agriculturally important crops (Leskey and Nielsen 2018). In our examination of the literature, there is currently no documentation of *H. halys* feeding on industrial hemp, *Cannabis sativa* L. (Lago and Stanford 1989, McPartland et al. 2000). Herein, we report our observations of this invasive stink bug on grain variety industrial hemp (*C. sativa*) in Virginia.

Methods and Results

In September of 2016, Thomas P. Kuhar inspected a research planting of industrial hemp at Virginia Tech's Kentland Farm in Whitethorne, VA (37.196106N, −80.580221W). At time of inspection, plants were mature with fully developed seed heads and numerous *H. halys* adults were observed feeding on seeds (Figure 4.1). Since initial observations in 2016, *H. halys* has remained the most commonly observed stink bug species on grain/fiber hemp at this location in 2017 and 2018. Nymphs, adults, and eggs of this species have been found on plants (Figures 4.2 and 4.3). On 28 August 2018, we received laboratory colony *H. halys* egg masses from USDA-ARS in Beltsville, MD. On 7 September 2018, we placed 28 second instars into a cage containing a potted *C. sativa* plant along with fresh field-harvested seed heads of *C. sativa*. Survival and development of *H. halys* was assessed comparatively against corn (*Zea mays L.*), a known suitable host plant (Kuhar et al. 2012); this was evaluated in four cages (*n* = 4) for each host plant type. The study was terminated on 8 October 2018 when there were no remaining live insects in cages. Nymphs successfully completed development on both *Z. mays* and *C. sativa* with an average of 24% (2, 10, 4, and 1 stink bugs developing to adult stage) and 66% (20, 23, 23, and 9 stink bugs developing to adult stage), respectively, which is similar, if not higher, to other published studies of *H. halys* development on various beans, seeds, carrot, or tree fruit (Nielsen et al. 2008, Medal et al. 2012, Acebes-Doria et al. 2016, Dingha and Jackai 2016); Nielsen et al. (2008) observed 52.5% of *H. halys* nymphs on a diet of beans and peanuts and Dingha and Jackai (2016) reported 60–80% survival of *H. halys* nymphs on carrot, green beans, princess tree leaves, and various seeds. Given the developmental success on *C. sativa* compared with *Z. mays*, it appears that *C. sativa* may be a suitable host plant for *H. halys*. More evaluations are needed on reproductive ability to understand the suitability of *C. sativa* as a host for *H. halys*.

Discussion and Conclusion

To date, no qualitative or quantitative effects of brown marmorated stink bug feeding injury have been observed on leaves, stems, or seeds of *C. sativa*. Among the various types of *C. sativa* crop plants, including those grown for fiber, cannabidiol oil, and marijuana, it is grain hemp that would likely be the most vulnerable to stink bug injury. *Halyomorpha halys* feeds on fruiting or reproductive portions of plants (Kuhar et al. 2012) and in *C. sativa*, seeds from grain varieties are the fruiting or reproductive portions. Occasionally, seeds with a hollow center (unviable seeds) are collected from plants, but at this time we are uncertain if this injury is caused by brown marmorated stink bugs. In 2018, a replicated field study was conducted in which *H. halys* nymphs were caged on grain hemp plants using 20-liter paint strainer bags at varying densities of 0, 10, and 20 stink bugs per developing seed head/flowering portion of the plant. The experiment was conducted at the aforementioned Kentland Farm and plant and insect health were assessed weekly. Although brown marmorated stink bugs were observed feeding upon seeds and flowering portions of the plant, seed weight was similar among treatments and there was no visually detectable reduction in quality of seeds. More work should be done to determine the effects of brown marmorated stink bugs on *C. sativa*. However, based on this research, it does not appear that *H. halys* poses a serious threat to industrial hemp.

Evaluation of insecticides

Hemp production in the U.S. increased dramatically in 2019, and still very little information is published with regards to pest management on the crop. The objective of our experiment was to assess the efficacy of several natural insecticide products for control of BMSB, which is a common pest of grain hemp in Virginia. A small plot field experiment was conducted on a planting of 'Felina 32' hemp direct seeded with a grain drill at 30 lb seed per acre on 31 May 2019 at the Virginia Tech Kentland Farm. The experiment had six treatments (BoteGHA (*Beauveria bassiana* strain GHA); Trilogy (clarified hydrophobic extract of neem oil);

Entrust (spinosad); Azera (azadirachtin and pyrethrins); Requiem (terpene constituents of the extract of *Chenopodium ambrosioides*)); and an untreated check arranged in an RCBD with four replicates. Individual plots were 6 ft × 10 ft surrounded by 5 ft alleys with no plants. Hemp plants were sprayed with products in the field using a single-nozzle boom with D3 spray tip powered by a CO_2 backpack sprayer set at 40 psi, and delivering 30 gal per acre. Treatments were applied on 13, 20, and 27 Aug. On 16, 23, and 30 Aug, five plants per plot were visually inspected for stink bugs. On 20 Aug., after allowing plants to dry for 4 hr in the field after application, one leaf and one seed head from each plot were excised, placed in a 1 qt plastic deli container, and brought back to the lab for a bioassay with live field-collected BMSB. Approximately 120 late-instar BMSB nymphs were collected from red bud and catalpa trees around Blacksburg, VA within 1 to 3 d prior to the bioassay and were held in a mesh cage with a water wick prior to testing. BMSB were placed 5 bugs per container and mortality was recorded at 1, 2, and 3 DAT. Data were analyzed using analysis of variance (ANOVA) procedures and means were separated using Fisher's LSD at the 0.05 level of significance.

BMSB densities were low on field plots and there was no significant effect of treatment. In the BMSB laboratory bioassay, a significant treatment effect occurred at 3 DAT, at which time, Azera had the highest percentage mortality at 60.0%, which was higher than all other treatments except Requiem (40.0%). BMSB mortality on plants treated with BoteGHA, Trilogy, and Entrust was not significantly different from the untreated check (Table 4.1).

Figure 4.1: Brown marmorated stink bug adult on *C. sativa*.

Figure 4.2: Brown marmorated stink bug nymph on *C. sativa*.

Figure 4.3: Brown marmorated stink bug eggs on *C. sativa*.

Table 4.1: Counts of brown marmorated stink bugs and mortality of bugs placed on treated foliage and seeds of field plots of hemp treated with various insecticides in Whitethorne, VA in 2019.

		BMSB adults or nymphs per five plants			BMSB mortality (%) on treated hemp leaves and seeds		
Treatment	fl oz/acre	Aug 16	Aug 23	Aug 30	1 DAT	2 DAT	3 DAT
Untreated check	---	0.0	0.3	0.3	5.0	12.5	17.5c
BoteGHA	37.2	0.3	0.3	0.3	5.0	17.5	30.0bc
Trilogy	44.1	0.3	1.0	0.5	0.0	735	17.5bc
Entrust	5.0	0.3	0.3	0.3	0.0	30.0	32.5bc
Azera	32.0	0.3	0.8	0.3	10.0	22.5	60.0a
Requiem	128.0	0.0	1.0	0.3	5.0	10.0	45.0ab
P		ns	ns	ns	ns	ns	<0.005

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Chapter 5: Defoliation effects on yield and cannabinoid content of hemp

Introduction

Hemp (*Cannabis sativa* L.) production in the U.S. has increased dramatically since full legalization of the crop through passage of the Agricultural Improvement Act of 2018 (U.S. H.R. 2 –115th Congress [115-334]). However, due to the long moratorium on *C. sativa* in the United States, there remains much to learn about crop production and management, including the pest complex and associated impacts. In the U.S., many leaf-chewing insect herbivores occur on hemp including orthopterans (various grasshoppers and crickets), coleopterans (Japanese beetle, *Popillia japonica* Newman; green June beetle, *Cotinis nitida* L.; spotted cucumber beetle, *Diabrotica undecimpunctata howardi* Barber; flea beetles, including *Psylliodes* spp., *Phyllotreta* spp., and *Podagrica* spp., etc.), and lepidopteran larvae (corn earworm, *Helicoverpa zea* [Boddie]; yellowstriped armyworm, *Spodoptera ornithogalli* [Guenée]; and bertha armyworm, *Mamestra configurata* Walker), among others (McPartland et al. 2000, Cranshaw et al. 2019). However, leaf chewing insect feeding injury impact on yield and cannabinoid production remains unknown (Cranshaw et al. 2019). Intuitively, any loss of leaf area should result in an immediate loss of photosynthetic capability at some level, but how plants respond to this stress is complex and varies across plant species and conditions (Wyatt et al. 1990, Obeso 1993, Iqbal et al. 2012). Overall, plants defend themselves against herbivores using two broad strategies: tolerance and resistance (Núñez-Farfán et al. 2007, Wise and Abrahamson 2007, Fornoni 2011, Mitchell et al. 2016).

Herbivores have long been associated with plants and the timing and amount of their defoliation has a great influence on plant reproductive success (Wyatt et al. 1990). Plants are more vulnerable to herbivores depending on the growth stage and in some, early season defoliation has a greater impact due to depleted nutrient reserves (Stuart Chapin III 1980). As a response to defoliation, many plants exhibit a tolerance for tissue loss (Crawley 1983). In perennial crops, impacts of defoliation can persist for several years (Tuomi et al. 1984). In oak trees, for example, defoliation by gypsy moth larvae leads to a decline in leaf quality (e.g., tougher texture, reduced water content), which reduces benefits to herbivores, leading to decreased feeding of leaves in subsequent years (Schultz and Baldwin 1982). Short-term compensation can be seen in some herbaceous perennials in response to severe leaf damage (Obeso 1993). In velvetleaf (*Abutilon theophrasti* Medik), plants defoliated at 75% resulted in reduced seed production when sown at a high density but experienced no negative impacts when sown at low density (Lee and Bazzaz 1980) as light was better able to penetrate the lower canopy (Mabry and Wayne 1997). Similarly, mustard defoliated early in the season led to increased leaf production and plant dry mass (Khan and Lone 2005).

In addition to tolerance traits, plants may also defend against herbivory with resistance traits that reduce the preference and/or performance of herbivores. Resistance traits, such as secondary metabolites, can be induced following damage. Unique to cannabis plants are cannabinoids, secondary metabolites produced in glandular trichomes found only in foliar material (Fairbairn 1972, Sirikantaramas et al. 2005, 2008), which are of major importance to the hemp industry. Several cannabinoid acids or alkaloids are reported as plant defense compounds (Sirikantaramas et al. 2005, Morimoto et al. 2007, Bidart-Bouzat and Imeh-

Nathaniel 2008) and, similarly, McPartland et al. (2000) reported that chewed hemp plants produce more secondary defensive chemicals, such as THC (tetrahydrocannabinol). These plant responses may reduce damage from herbivores, but are also a serious concern as the current state and federal legal threshold for THC allowance in hemp plants is not to exceed 0.3%.

The aim of this study was to determine the effects of defoliation at various levels and timings on yield and cannabinoid concentrations of hemp grown for grain and cannabinoid production in Virginia. This information will contribute to our understanding of how plant-pest interactions impact yield and quality within this cropping system and will ultimately aid our pest management decision-making.

Materials and Methods

Grain hemp experiment, 2018 and 2019

Experiments were conducted at Virginia Tech's Kentland Research Farm in Whitethorne, VA (37.194664, -80.577227), where plots of 'Felina 32' dual-purpose (grain/fiber) hemp were planted on 8 June 2018 and 31 May 2019. Hemp was direct-seeded at a rate of 33.6 kg per ha with a grain drill both years. In 2018, a pre-emergence herbicide containing glyphosate (41%) (Cornerstone Plus, Winfield Solutions, LLC, St. Paul, MN) was applied to manage weeds (2.4 kg of product per hectare) and in 2019, weeds were managed prior to planting using a flame burndown. Treatments included four defoliation amounts (0, 25%, 50%, and 75% leaf removal) and three defoliation timings (20-, 40-, and 60-days post planting). Every leaf on every plant in a plot was defoliated manually using hand-held shears (Fiskars® Micro-Tip® Pruning Snips, Helsinki, Finland). Similar to methodology used by Cranshaw and Radcliffe (1980) in potatoes, defoliation events were instantaneous, incorporating one amount of defoliation at one timing. Field plots were arranged in a split-plot design with defoliation timing as a whole-plot factor and defoliation amount as a sub-plot factor, both assigned in a randomized complete block design. In 2018, individual sub-plots were 1.2 m × 1.2 m (approximately 40 plants) and all treatments were replicated in four sub-plots. In 2019, plot sizes were approximately 0.6 m × 0.6 m (approximately 15 plants) and all treatments were replicated in eight sub-plots. In both years between days 20 and 60, a broad-spectrum insecticide containing two active ingredients, lambda-cyhalothrin (4.63%) and chlorantraniliprole (9.23%) (Besiege, Syngenta Crop Protection, Greensboro, NC), was applied (658 ml of product per hectare) every ten days using a CO_2 backpack sprayer to prevent naturally-occurring insect herbivory.

Plants were hand harvested 90 days after planting. Plant material was stored in paper bags and transported to the laboratory for analysis. Seeds and seed head floral and foliar material were shucked away from stems and a sieve was used to separate the material. Seed weight was taken in bulk for all plants harvested from a sub-plot and data were recorded as average seed weight per plant for each sub-plot. Seed head foliar material was placed into paper bags and stored in a -80°F freezer for preservation until cannabinoid analysis via high-performance liquid chromatography (HPLC). Samples from both years were removed from the -80°F freezer on the same day and taken to a commercial cannabis testing lab (ECC Test Lab, Blacksburg, VA) for HPLC analysis. Yield and cannabinoid data were analyzed separately by year via two-way ANOVA (JMP® Pro 15, SAS Institute Inc., Cary, NC) to assess differences due to defoliation timing, defoliation amount, and the interaction of both treatments on yield (average seed weight per plant), cannabidiol (CBD) content, and tetrahydrocannabinol (THC) content. In cases of significant interactions between defoliation amount and defoliation timing, data were split by timing and analyzed separately via one-way ANOVA. Yield data were not normally distributed and were square root transformed to improve normality. THC data were zero-inflated and were not normally distributed, therefore zero values were dropped from the dataset, and 2018 and 2019 values were merged for analysis and arcsine square root transformed to improve normality.

Cannabinoid hemp experiment, 2019 and 2020

Experiments were conducted at Virginia Tech's Catawba Sustainability Center in Catawba, VA (37.384706, -80.095099), where seedlings of the CBD hemp cultivars 'Spectrum' (in 2019) and 'Wife' and 'BaOx' (in 2020) were transplanted on 2 July in both years. Seedlings were transplanted into raised beds covered with white polyethylene mulch that were drip irrigated approximately every 14 days. Plants were defoliated 50% at 20-, 40-, or 60-days post-transplanting, in addition to control plants that were not defoliated. Field plots were arranged in a randomized complete block design with four replicates. As CBD hemp plants are quite large, a single plant was used as a plot. The same shears described in the previous experiment were used to manually remove 50% leaf tissue area from every leaf per plant.

Approximately 90 days after transplanting, whole plants were hand harvested and hung in a barn to dry for approximately 28 days. After drying, buds were stripped from plants and total yield, as well as a 100-bud weight, were recorded for each plant. Bud material (4 per plant to make a composite sample) was collected prior to harvest, placed in paper bags, immediately stored on ice in a cooler for transport to the lab, then stored in a -80^{o} freezer for preservation until chemical analysis. All 2019 and 2020 samples were removed from the -80^{o}F freezer on the same day and taken to a commercial cannabis testing lab (ECC Test Lab, Blacksburg, VA) for HPLC analysis. Data were separated by variety and analyzed using one-way ANOVA (JMP® Pro 15, SAS Institute Inc., Cary, NC) to assess effects of defoliation timing on cannabidiol (CBD) content, tetrahydrocannabinol (THC) content, total bud weight per plant, and 100 bud weight per plant. Total and 100 bud weight data for Wife and BaOx cultivars were not normally distributed, so data were square root transformed to improve normality.

Results

Grain hemp experiment, 2018 and 2019

No significant treatment or interaction effects were found for seed yield in either year or overall (Figures 5.1 and 5.2), total CBD in 2019 (Figure 5.3), or total THC (Figure 5.4). For 2018 CBD content, there was a significant interactive effect of defoliation timing and defoliation amount in relation to CBD content ($p = 0.01$); specifically, at 40 days post planting, plants defoliated 50% had significantly more CBD than plants defoliated 75%. In 2018, CBD data were grouped by defoliation timing and analyzed via one-way ANOVA and there were no significant differences. Although there were no effects of defoliation on THC content, it should be noted that cannabinoid contents in grain varieties were miniscule and values were not close to exceeding the 0.3% legal threshold.

Cannabinoid hemp experiment, 2019 and 2020

Variety had a significant effect on bud yield and cannabinoid content and, thus, defoliation treatments were analyzed separately by variety. In all varieties, defoliation timing had no effect on CBD content (Figure 5.5), THC content (Figure 5.6), total bud weight per plant (Figure 5.7), or 100 bud weight per plant (Figure 5.8). Although there was only one level of defoliation applied to plants in this experiment (50%), results indicate that defoliation likely does not impact physical crop yield (bud weight) or cannabinoid content. THC content in all varieties exceeded the legally allowed 0.3% threshold, but this can be attributed to varietal differences and was not a result of treatment effects.

Discussion

This study simulated chewing insect herbivory on foliar surface area of grain and cannabinoid hemp cultivars at various times and amounts to determine impacts on yield and cannabinoid production. Findings indicated that over two growing seasons in Virginia, manual removal of leaf tissue of up to 75% in grain and 50% in CBD cultivars at either 20, 40, or 60 days after planting did not significantly impact observable effects on physical yield (seed or bud weight) or cannabinoid content (CBD or THC) at time of harvest. Thus, the tolerance and resiliency of hemp to defoliation appears to be consistent across both grain and CBD cultivars.

Experimental results suggest that hemp may have a higher tolerance to environmental stressors that impact leaf surface area, or that defoliation at the levels applied does not cause enough stress to affect yield or cannabinoid production in the varieties tested. This is in contrast to many other annual crops; depending upon timing and severity, leaf removal has been shown to significantly reduce yield of numerous crops including wheat (Sharrow 1990), sugar beet (Muro et al. 1998a), onion (Muro et al. 1998b), garlic (Muro et al. 2000), sunflower (Muro et al. 2001), corn (Lauer et al. 2004), soybean (Todd and Morgan 1972, Hammond and Pedigo 1982, Hammond 1989, Hunt et al. 1994, 1995), snap bean (Greene and Minnick 1967), and cowpea (Ibrahim et al. 2010). In the grain and CBD cultivars used for this experiment, yield and cannabinoid concentrations remained stable regardless of the amount or timing of defoliation. This experiment did not test immediate effects to cannabinoids at time of defoliation; however, if cannabinoids are impacted at the time of defoliation, this experiment shows that values likely stabilize by time of harvest, which is consistent with the timing that producers are subjected to testing by state departments of agriculture. Observations in Virginia indicate that natural defoliation of hemp rarely exceeds 25%; these observations are representative of a single leaf on a plant, not a full plant defoliation as tested in these experiments. Thus, the treatments selected for this experiment were equal to or much greater than any level of naturally-occurring defoliation observed and yet still did not appear to impact yield or cannabinoid concentrations.

It should be noted that response may have been different if the damage inflicted was the result of true insect herbivory due to insect saliva; particularly salivary injections by insects at time of feeding (Quentin et al. 2010), as certain components have been shown to impact plant response (Steinbauer et al. 1997). However, while plant response to artificial defoliation may be not be as pronounced as true defoliation, artificial defoliation studies are usually a good indicator of plant response to natural injury (Todd and Morgan 1972, Hammond and Pedigo 1982, Quentin et al. 2010). Much of the observed insect presence in hemp is not limited to leaf surfaces; insects are also present and feed from crop reproductive structures, such as seeds (in grain cultivars) and buds (in CBD cultivars) and it is likely that impacts to yield and cannabinoid production would be greater if these plant tissues are fed upon. Additionally, secondary metabolites such as cannabinoids (Fairbairn 1972, Sirikantaramas et al. 2005, 2008) can have a role in plant physiology including defense, growth, and development (Kutchan 2001). Since cannabinoid content was not elevated in response to leaf damage, it is likely that the plant can tolerate the chosen levels of defoliation for this study or that defoliation treatments were not severe enough to elicit a response.

This experiment indicates that final yield or cannabinoid content in hemp is not impacted by a loss of leaf surface area and, in the context of chewing insect pests, management may not be warranted. These findings may have implications beyond chewing insects as plant defoliation can occur from a number of biotic and abiotic factors, such as invertebrate and vertebrate herbivores, plant pathogens, hail, etc. In addition, research in other crops also indicates that growing conditions and other environmental factors can impact a crop's response to defoliation (Caviness and Thomas 1980, Hunt et al. 1994). Regardless of the stressor, results of this study suggest that hemp may be generally quite tolerant to leaf surface area loss. We encourage future defoliation experiments in hemp to 1) occur at multiple field sites, 2) include different crop varieties, 3) assess impacts from multiple defoliation events at more than one instantaneous point throughout the season, 4) assess cannabinoid responses at more times throughout the season than just harvest, and 5) assess impacts of simulated or true insect herbivory on hemp reproductive structures such as seeds or buds.

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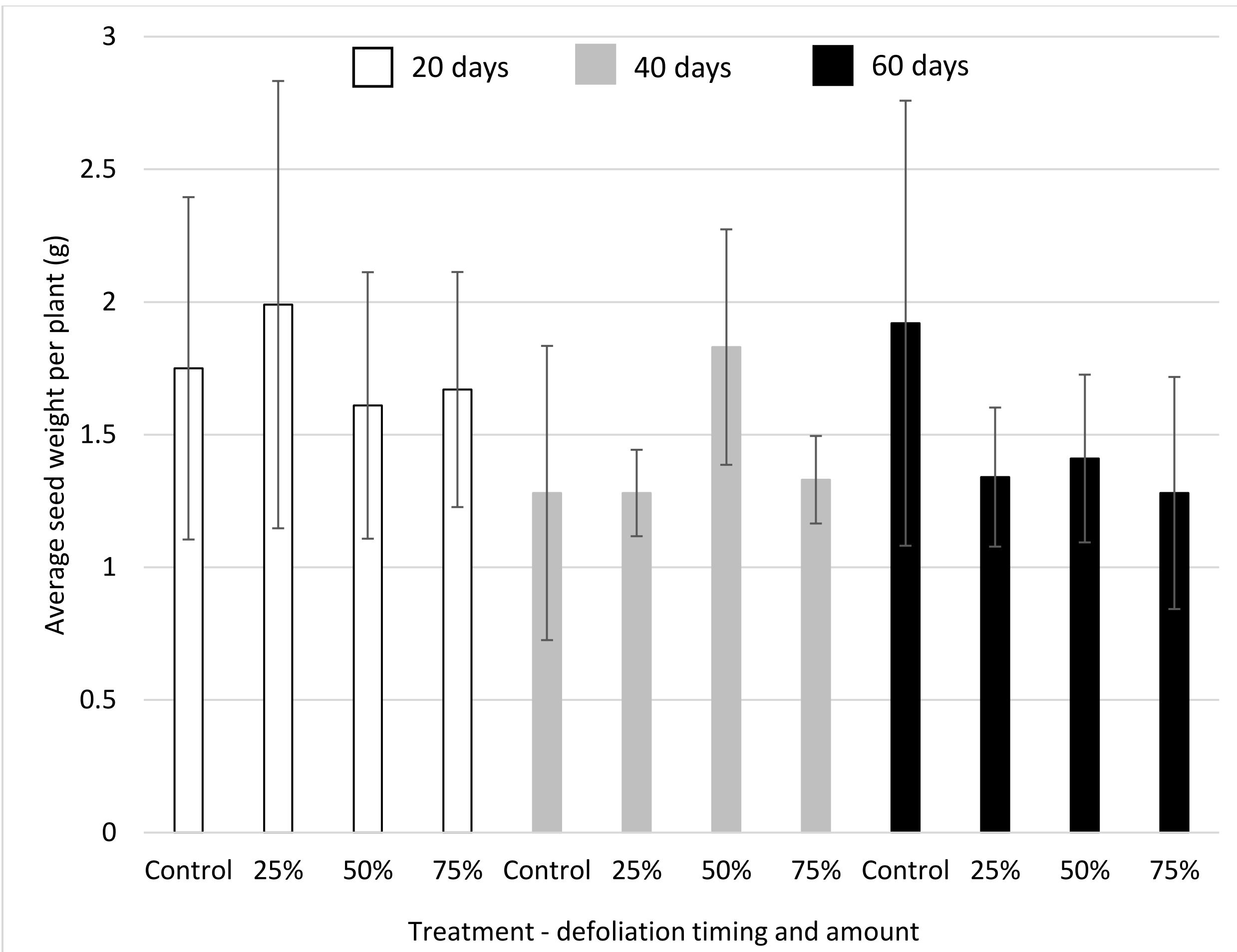
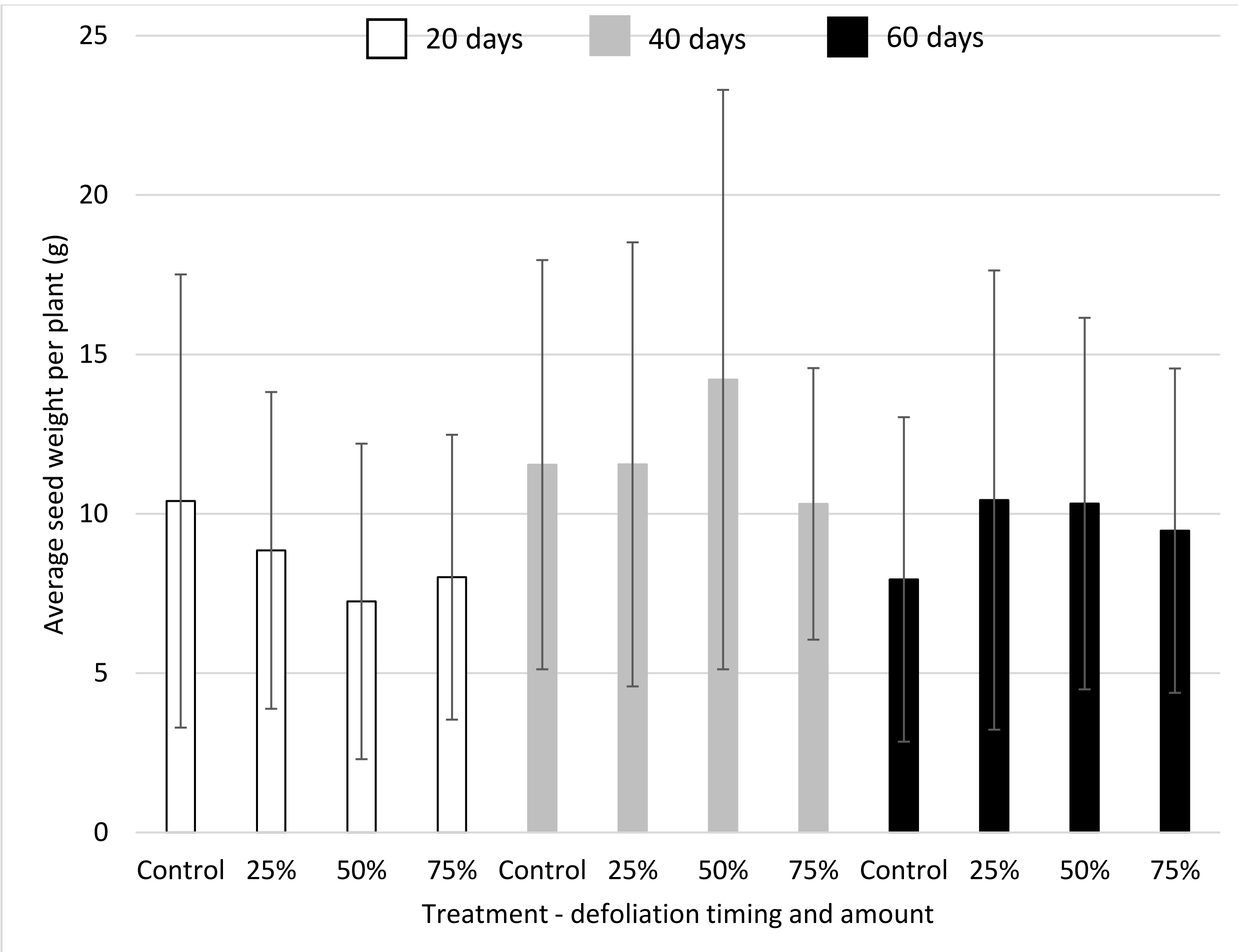
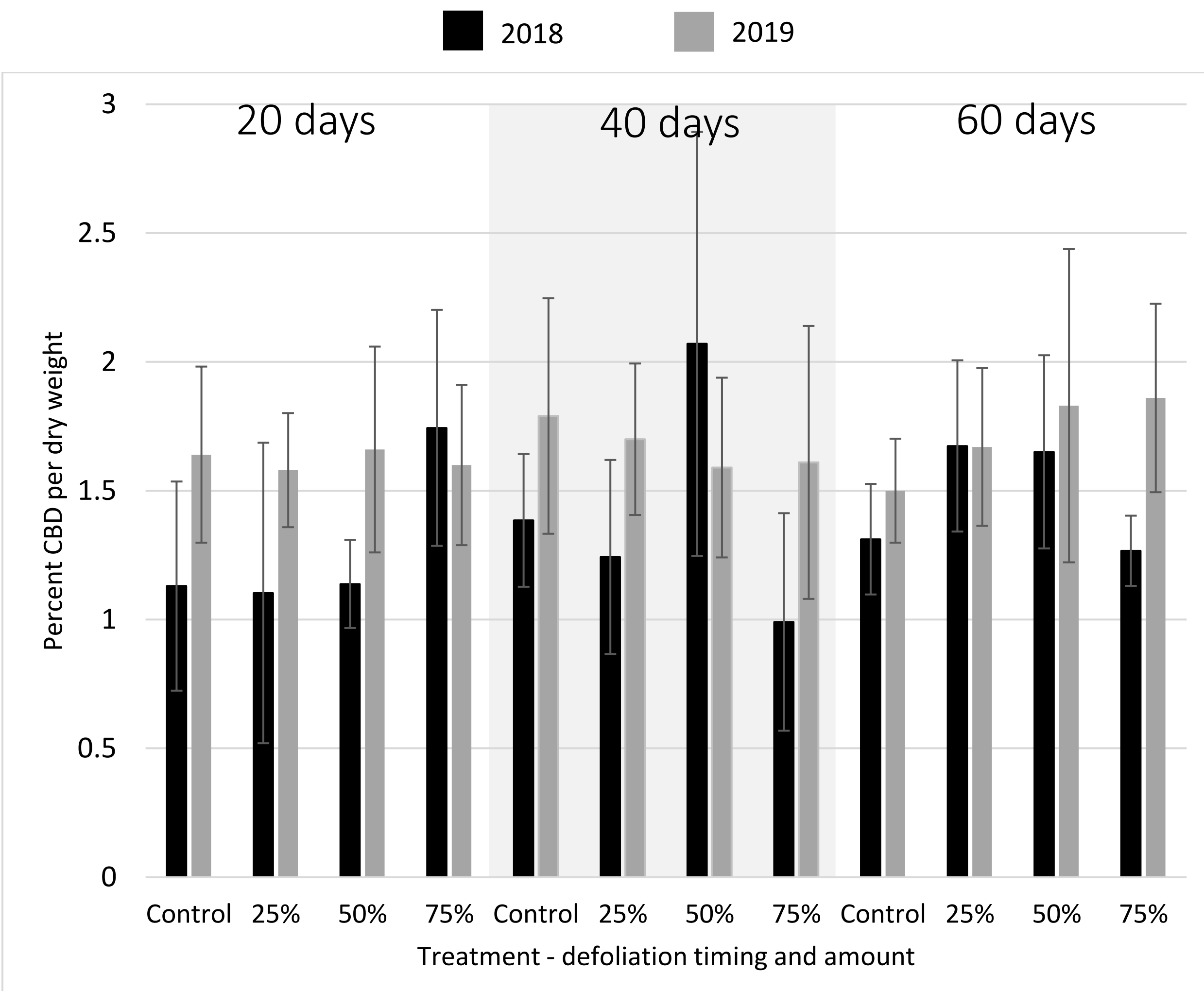
Figure 5.1: 2018 grain hemp yield. Data were square root transformed to improve normality and analyzed via two-way ANOVA. Defoliation time, p = 0.20; defoliation amount, p = 0.76; defoliation time*defoliation amount, p = 0.38. Graph provides means +/- standard deviation across N=4 replicate plots.



<u>Figure 5.2</u>: 2019 grain hemp yield. Data were square root transformed to improve normality and analyzed via two-way ANOVA. Defoliation time, $p = 0.70$; defoliation amount, $p = 0.92$; defoliation time*defoliation amount, $p = 0.95$. Graph provides means +/- standard deviation across N=8 replicate plots.



<u>Figure 5.3</u>: 2018 and 2019 grain hemp cannabidiol (CBD) content. Data were analyzed via two-way ANOVA (defoliation timing, defoliation amount, and interaction). Analysis of 2018 data showed a significant interaction effect (p = 0.01), so data were grouped by defoliation timing and analyzed via one-way ANOVA (20 days, p = 0.16; 40 days, p = 0.06; 60 days, p = 0.15). There were no significant treatment effects in 2019 (Defoliation time, p = 0.73; defoliation amount, p = 0.95; defoliation time*defoliation amount, p = 0.46). Graph provides means +/- standard deviation across N=4 (2018) and 8 (2019) replicate plots.



<u>Figure 5.4</u>: 2018 and 2019 grain hemp tetrahydrocannabinol (THC) content. Data from both years were merged, arcsine square root transformed to improve normality, and analyzed via two-way ANOVA. Defoliation time, p = 0.78; defoliation amount, p = 0.11; defoliation time*defoliation amount, p = 0.44. Graph provides means +/- standard deviation across N=22 (2018) and 42 (2019) samples.

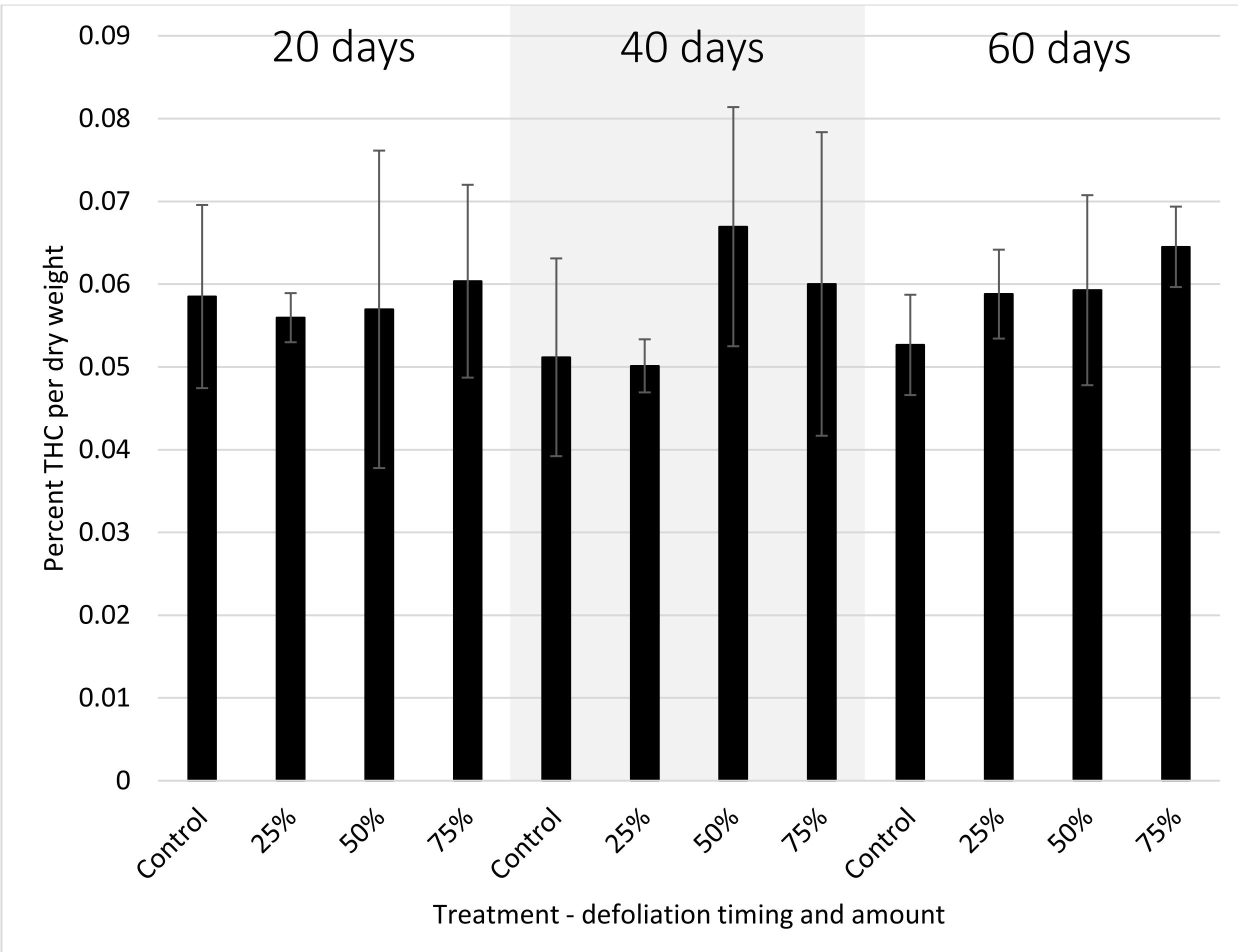
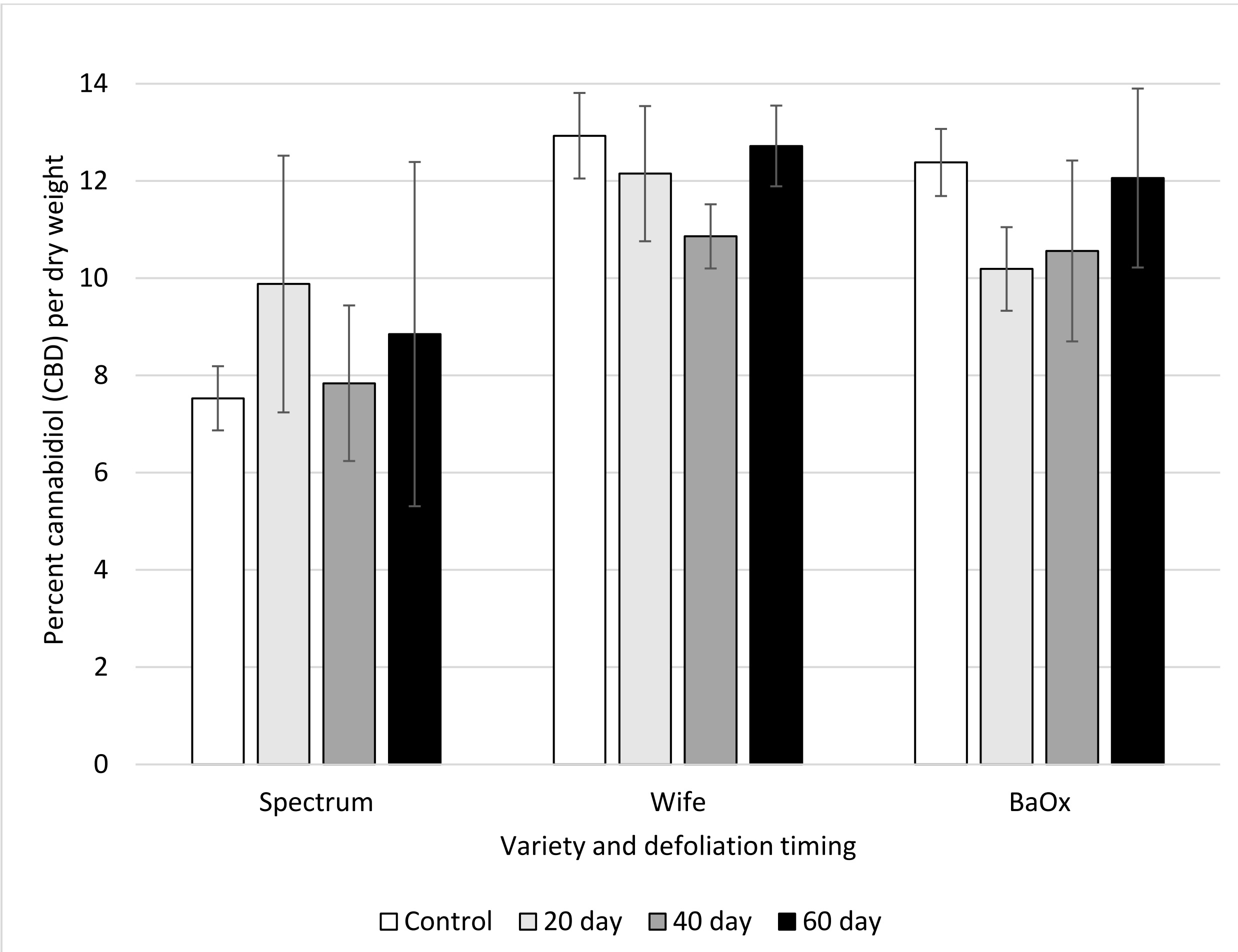


Figure 5.5: Cannabidiol (CBD) content in CBD hemp cultivars. Data were analyzed via one-way ANOVA to assess impact of defoliation timing. Spectrum p = 0.73; Wife p = 0.17; BaOx p = 0.21. Graph provides means +/- standard deviation across N=4 replicate plots.



<u>Figure 5.6</u>: Tetrahydrocannabinol (THC) content in CBD hemp cultivars. Data were analyzed via one-way ANOVA to assess impact of defoliation timing. Spectrum p = 0.96; Wife p = 0.21; BaOx p = 0.13. Graph provides means +/- standard deviation across N=4 replicate plots.

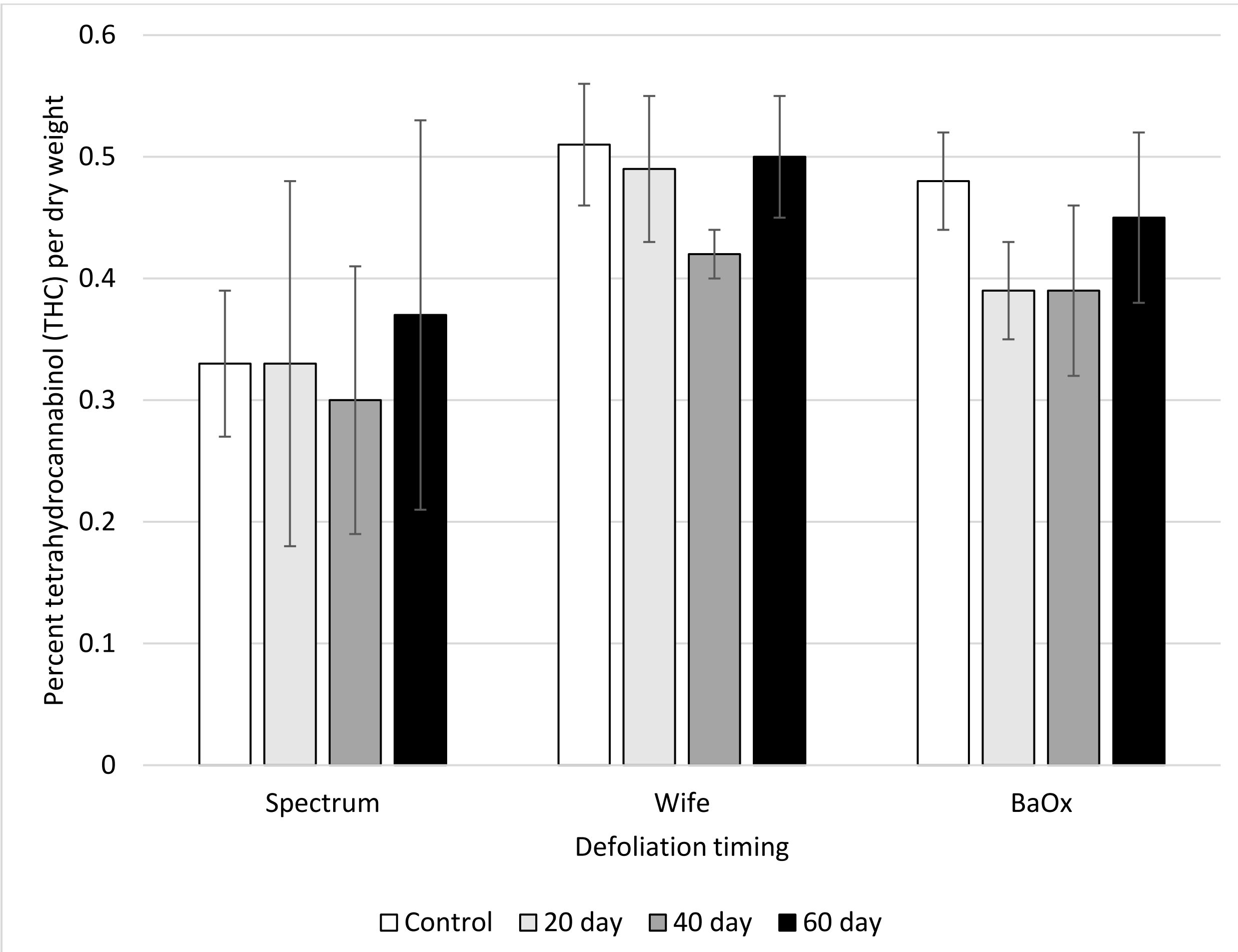


Figure 5.7: Total weight (g) of all buds per plant in CBD hemp cultivars. Wife and BaOx data were square root transformed to improve normality. Data were split by variety and analyzed via one-way ANOVA to assess impact of defoliation timing. Spectrum p = 0.99; Wife p = 0.36; BaOx p = 0.93. Graph provides means +/- standard deviation across N=4 replicate plots.

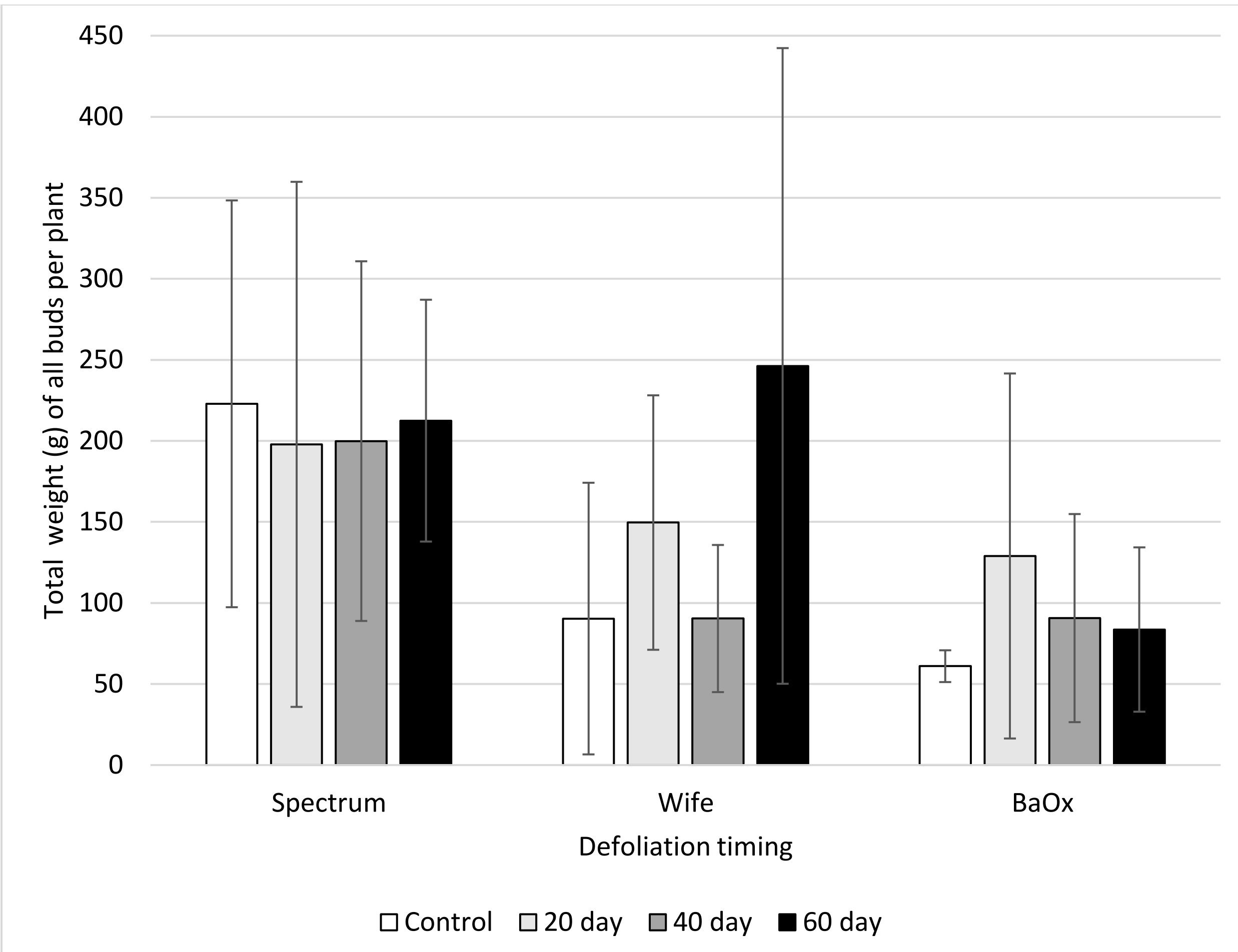
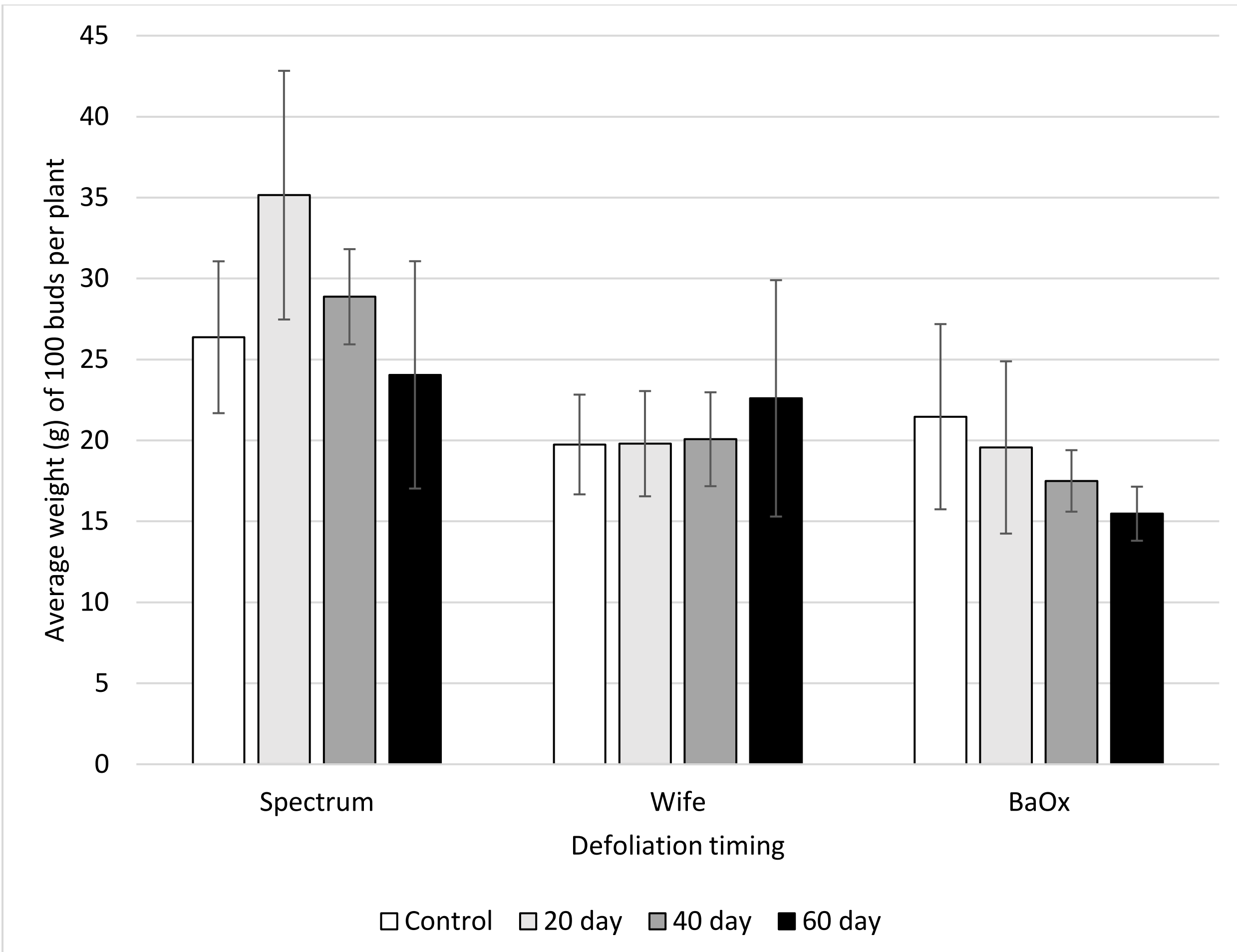


Figure 5.8: 100 bud weight (g) in CBD hemp cultivars. Wife and BaOx data were square root transformed to improve normality. Data were split by variety and analyzed via one-way ANOVA to assess impact of defoliation timing. Spectrum p = 0.36; Wife p = 0.88; BaOx p = 0.94. Graph provides means +/- standard deviation across N=4 replicate plots.



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Conclusion

This work has been some of the first to document insect pest presence and associated impacts to hemp in the United States. Specific outputs of this project include 1) established information in regards to corn earworm's association with outdoor produced hemp in Virginia, including evaluation of specific monitoring and management programs and evaluation of insecticides in a 2) lab and 3) field setting, 4) evaluation of hemp as a host plant and feeding impacts of brown marmorated stink bug, and 5) documented effects of foliar area loss on yield and cannabinoid production in grain and cannabinoid hemp varieties.

Corn earworm is a highly injurious pest to hemp produced outdoors in all locations of the United States and Virginia hemp has not been spared. This research has provided greater details in regards to the timing of corn earworm presence in hemp and appearance of crop injury as a result of chewing behavior. Evaluation of monitoring techniques has shown that the use of pheromone traps may be valuable in determining when corn earworm moths are present in the vicinity of hemp fields, but is not useful in predicting larval presence in buds or final crop damage. There is a relationship between larval presence and final crop damage and future studies should explore this further. Several insecticides are currently available and allowed for corn earworm management in hemp and were assessed for efficacy. Products containing *Bacillus thuringiensis* varieties *kurstaki* or *aizawai* only offer moderate efficacy against corn earworm in field settings. Products containing polyhedral occlusion bodies of *Helicoverpa zea* nucleopolyhedrovirus offer slightly better efficacy in field settings but more research should occur to assess application frequency and epizootic effects in hemp. Corn earworm management in hemp will remain a challenge due to insecticide limitations and lack of effective monitoring tools. As it stands, hemp must be scouted and monitored on a weekly (or similarly consistent) basis once flowering begins. Once corn earworm larvae are detected in buds, management tactics should be initiated.

Brown marmorated stink bug is a highly injurious pest to many crops and it has been observed in outdoor Virginia hemp all seasons thus far. There was concern of similar damage occurring in hemp, so studies were initiated to determine the potential of hemp as a host plant and potential resulting crop injury from brown marmorated stink bug feeding. Results showed that brown marmorated stink bug can survive and develop exclusively on hemp but no feeding injury could be detected after one week of caging nymphs and adults on grain hemp seed heads in the field. Information about this pest in hemp has been extremely useful, but future studies should occur with other species of stink bugs and stink bug feeding effects on cannabinoid content in plants should be explored.

Defoliation studies showed no observable effects on yield (seed weight in grain and bud weight in cannabinoid varieties) or cannabinoid content (specifically CBD and THC) in grain (one variety) and cannabinoid hemp (three varieties) in Virginia. This information would be valuable at any stage but is particularly helpful during the early period of hemp cultivation in the United States. Information can now be shared with growers that loss of hemp foliar area at any point during the growing season will not be a cause for concern and management of leaf-feeding pests is likely not warranted. However, this work should be modified and expanded in the future to explore additional parameters of insect defoliation in hemp.

The moratorium on hemp production and research for such an extended period of time has greatly complicated the ability to document valuable and needed information on a crop that is currently receiving such widespread attention. Every research question addressed here has created more targeted questions to be explored in future studies. As research continues in the coming years, pest monitoring and management strategies will become more established.

The role of hemp in United States agriculture is still being determined. Hemp occupies a unique, niche space (multiple crops in one plant – food, fiber, medicine), but the stability of the commodity remains to be seen. Greater infrastructure and capital investment are needed to aid long-term success. High-THC varieties of *Cannabis sativa* may have a more permanent role and by seizing the opportunity to conduct research with cannabinoid hemp cultivars, we are better prepared to address future pest issues as they arise.

Appendix I: Insect and mite pest management in hemp

As published in**: Britt, K., J. Fike, M. Flessner, C. Johnson, T. Kuhar, T. McCoy, and T. D. Reed**. **2020**. Integrated pest management of hemp in Virginia. Virginia Coop. Ext. Publ. No. ENTO-349. 29 pp.

A wide diversity of insects and mites can be found on hemp and many of these are discussed extensively in a recent open-access publication (Cranshaw et al. 2019). Some insects and mites are generalists that come to hemp opportunistically to feed, such as corn earworm and spider mites. These insects and mites feed on a wide variety of plant species, including hemp. Because of this, it is likely that many of the insects and mites seen in hemp are incidental opportunists. Others, such as cannabis aphid and hemp russet mite, appear to be specialists that require cannabis to survive. The vast majority of insects and mites observed on the plant do not appear to be significant pests based on our work over the past three growing seasons (2017-19). A few species certainly have proven to be important pests and these will be discussed.

Insect and mite pests can be broadly categorized by their feeding behavior as either chewing or piercing-sucking types. **Chewing** pests damage plants by consuming foliage, flower, and/or seed material with mouthparts much like our own. They chew and devour portions of hemp plants, leaves, stems, flowers, or seeds. Beetles and caterpillars fall into this group. **Piercing-sucking** pests have syringe-like mouthparts that are used to pierce the plant. These insects feed from the plant's vascular fluids or they liquefy and suck out plant tissue. Because they do not chew holes in plant material, their injury to plants is not always discernible. Aphids and mites fall into this group.

When strategizing how to appropriately manage insect and mite pests in a crop system, it is best to utilize multiple management tactics via integrated pest management (IPM). One of the key elements of IPM is the establishment of economic thresholds for insect or mite injury to plants. By relying upon evidence-based economic thresholds, growers can address pest problems before they become a financial burden. Virginia hemp presents a unique pest management challenge because the crop has not been grown long enough for research to be done to establish the injury levels at which economic loss can occur. Although we have not yet established the important economic information that will help with managing pests, we have identified several insects that are likely to be problematic in hemp and are beginning to conduct studies to address the injury and damage that can result from their presence and feeding. Hemp does appear to be a more robust crop than others and can withstand a considerable amount of insect and mite feeding before crop injury occurs. However, it is certainly not exempt from insect and mite pest injury.

Hemp clones (rooted cuttings taken from mother plants) and seedling starts commonly are purchased for transplanting into the field in hemp flower production systems. Before introducing these transplants into your greenhouse or planting in a field, they should be briefly quarantined to prevent accidental pest introductions. Transplants often come with pests already present on plant material, such as aphids or mites. These pests can reproduce rapidly and once a population is established in a confined area, such as a greenhouse or warehouse, they are extremely difficult to manage or eradicate. Inspecting plants thoroughly prior to planting is an important preventative step in managing pests.

The following section outlines a few important insect and mite pests in hemp. At the end of this chapter, there are color photos to supplement the text.

Major insect and mite pests in hemp in Virginia

Corn earworm, *Helicoverpa zea*

Without a doubt, corn earworm is the most damaging pest of hemp grown in outdoor environments as it targets the marketable portions of hemp plants – floral regions of CBD and seeds of grain variety hemp. Corn earworm is a generalist chewing pest that feeds on a variety of economically important crops. Hemp is attractive to this insect and is a late season source of sustenance after most other crops have been harvested. Even though we have yet to establish economic thresholds and injury levels for corn earworm in hemp, this insect has caused economic loss for hemp growers. Corn earworm feeding and injury to hemp plants has most frequently been associated with elevated occurrences of bud rot on floral portions of CBD hemp (**color photo 1**). Bud rot results from the grey mold pathogen *Botrytis cinerea.* Corn earworm feeding does not directly cause bud rot; rather, the wounds on plants as a result of its feeding activity allow this opportunistic pathogen to invade and infect plants.

Corn earworm moths (**color photo 2**) have two periods of migration north to Virginia from more southern areas. In Virginia, corn earworm flights occur from mid-July to late August and during this time, female moths actively lay eggs in host crops. Cream-colored, spherical eggs are laid singly on plants. In hemp, eggs can typically be found on younger tissue in the flower bud or seed head. Neonate larvae emerge from eggs in 3 to 4 days and immediately begin feeding on plant material. Eggs and freshly-hatched larvae are so small in size that they typically go unnoticed. As larvae grow and develop, their coloration may change. Younger larvae are typically dark in coloration with prominent black bristles (**color photo 3**). Later stage larvae can vary in color from pink, yellow, green, brown, or even two-toned (**color photo 4**).

Larvae molt (or develop) through 6 to 8 instars (or growth stages) in a 2 to 3-week period, depending on environmental temperature; growth can be accelerated in areas with warmer climates. When fully developed, larvae drop from plants to the ground and burrow into soil to pupate (or develop into an adult moth). Pupae are dark red to brown in coloration. This insect overwinters (or goes into a type of hibernation) as a pupa. In Virginia, many generations of corn earworm can be expected during a growing season. In most areas of Virginia and states further north, relatively few pupae survive the winter due to low temperatures; however, it is possible for corn earworm to successfully overwinter in warmer areas of Virginia.

Regular scouting of hemp is important so that this insect can be spotted early and when worms are young. In some cases, environmental biological control will occur by organisms such as parasitic flies, spiders, predatory stink bugs, or pathogens. Hemp seems to be so attractive to corn earworm that it will preferentially lay eggs and feed on hemp regardless of the surrounding crops. In terms of chemical management, options are extremely limited. Of the few products currently approved for use on hemp in Virginia, those containing the active ingredient *Bacillus thuringiensis kurstaki* or *Bacillus thuringiensis aizawai* (Bt) are the best options for managing this insect (see pesticide table in Chapter 5). These products have selective activity against worms or caterpillars. If the choice is made to use Bt products, it is best to apply at the first sight of corn earworms as young worms are more susceptible to this product. Keep in mind that good plant coverage with Bt products is important because the worm must ingest the active ingredient to be killed. Corn earworm mortality from Bt will not be instant, but rather will occur after a few days. In 2019, efficacy trials conducted in Virginia showed that the Bt *aizawai* strain (found in the product XenTari) provided the best control among Bt products. The product Gemstar LC can also aid in reducing corn earworm infestations. Gemstar contains occlusion bodies of a nuclear polyhedrosis virus that is specific to only corn earworm.

Hemp russet mite, *Aculops cannabicola*

Hemp russet mite is perhaps the most injurious pest to indoor-grown hemp and is near impossible to eradicate once populations have established. This mite can attack outdoor plants, but in these cases, populations are usually more scattered and may impact only a few plants throughout a field. In indoor environments, every plant can quickly succumb to hemp russet mite injury. This mite is extremely small and is not visible to the naked eye. For perspective, hemp russet mite is less than half the size of twospotted spider mite. Additionally, multiple hemp russet mites can fit on the body surface of aphids. Hemp russet mites have four legs on their white- to beige-colored, cigar shaped bodies. Mites can only be seen under magnification (**color photo 5**) – microscopes are sufficient but hand lenses used to inspect hemp usually are not strong enough. Hemp russet mite is not easily managed and, due to its extremely small size, populations can quickly get out of control. Mites feed primarily on leaves, petioles, and shoot tips of hemp plants. Because hemp russet mite does not produce webbing on plants, its presence usually goes unnoticed until plants exhibit physical symptoms of stress. Advanced symptoms from extremely high populations can include upward curling of leaf edges (**color photo 6**), bronzing/russeting of leaf tissue (**color photo 7**), or a brown/tan powder appearance on leaf edges and stems which is actually an extremely heavy mite infestation. By the time plants express physical symptoms, irreparable damage to plants has already occurred.

There are few legal options available to manage hemp russet mite. Of the chemical products that we have tested, Venerate (Marrone Bio Innovations) has had the greatest efficacy. Current commercially-available predatory mites or other natural predators have not shown any promise at managing hemp russet mite. Prevention is the best tactic. Regular scouting of plants is a necessity. If any plants show signs of hemp russet mite, they should be moved away from other plants and quarantined until an accurate diagnosis can be made. Since hemp russet mite is so small, it is highly mobile and can easily spread throughout plants in close proximity. This is particularly a problem with indoor grow operations where fans are used to improve air flow.

Cannabis aphid, *Phorodon cannabis*

Cannabis aphid is a specialist, piercing-sucking insect that feeds exclusively on hemp. They can reproduce asexually, so populations can rapidly increase in favorable environments (**color photo 8**). Although these insects feed on sap from hemp plants, thus far, we have not observed direct plant injury by cannabis aphid presence or feeding. However, this insect excretes excessive amounts of honeydew (sticky, sugary waste) which creates sticky surfaces on plants. As aphids grow and molt (shed skin to grow), their shed skins can get caught in the sticky honeydew that remains on plant surfaces (**color photo 9**); this is undesirable for consumers of raw plant material. Honeydew is also an excellent substrate for sooty mold growth. The honeydew and sooty mold both present contamination issues in the raw plant material and mechanical issues during plant processing for cannabinoid extraction.

On outdoor plants, cannabis aphid eggs are laid in late fall. Overwintering eggs survive on crop debris and volunteer seedlings that sprout in fields the following spring will be colonized shortly after egg hatch occurs. At the end of the growing season, crop residue remaining in fields should be destroyed via tillage, burning, or another comparable method. This will aid in getting rid of any eggs that may have been laid in the late fall period. If eggs still happen to be present and aphids hatch out in early spring, there will be a reduced chance for survival if there is no leftover plant material for them to develop and feed on. Tillage helps, especially in grain hemp fields where seed shatter and dispersal occurs quite frequently and volunteer plants are most likely to be present.

Cannabis aphid is a much greater concern for indoor or greenhouse hemp production, especially in operations with perpetual harvests. These insects often are present on transplants that are acquired from indoor propagation. If you are growing plants to distribute to other growers, inspect plants prior to sale to make sure that they are clean and free of aphids. If you are receiving plants from an indoor facility, inspect your plants before planting them in the field or moving into your greenhouse. If planting indoors, it is of the utmost importance to quarantine the plants to ensure that they are free of aphids. If cannabis aphid is observed on any indoor plants, those plants should be removed from close proximity to other plants. Moving infested plants at least 10 feet or more away from clean plants would be a good start. Promptly addressing a few problem plants is far easier than attempting to control a widespread infestation, even if some plants must be destroyed. If this insect establishes a population indoors, it can be problematic and difficult to manage.

Outdoors, this insect is less of a concern due to the presence of natural enemies and environmental conditions. Predators, such as lady beetles (**color photo 10**), help manage aphid populations naturally and are excellent at finding aphid infestations to consume. Additionally, sufficient rainfall or dew aids in the removal of sticky honeydew from plant surfaces and helps prevent sooty mold problems. However, aphid outbreaks can occur and may require short-term mitigation in order to prevent excess sticky honeydew deposition on plants. Fortunately, a wide range of insecticides are available that can help reduce aphid numbers. These include pyrethrins, insecticidal soaps, neem oil, azadirachtin, and the bacteria-derived products Grandevo and Venerate (see pesticide table in Chapter 5). The risk of honeydew contamination is greater indoors since there is no natural rainfall to aid in removing the sticky substance. Predatory insects typically must be purchased for use in indoor growing environments because these insects do not typically live in such environments. Lady beetles are a feasible option for indoor use since the larval stage does not fly and adults are less likely to fly away from the crop. Adult lady beetles are not recommended for release in field settings as they are more likely to fly away in outdoor environments, but supplemental eggs or larvae may help provide some aphid management.

Twospotted spider mite, *Tetranychus urticae*

Twospotted spider mite is a generalist, piercing-sucking mite pest usually found on the undersides of plant leaves. Feeding injury to plants causes white stippling marks on leaves (**color photo 11**). This mite is small and oval in shape, has 8 legs, and can be orange/red or brown with two distinct dark spots on the body (**color photo 12**). Mites can be seen with the naked eye but microscopy can assist with proper identification. When mite populations are extremely high, webbing can be observed on leaves or in hemp flower buds or seed heads.

Twospotted spider mite is a concern for indoor hemp production. This mite thrives in dry, arid environments and, given the high humidity levels throughout Virginia, plants grown outdoors are not as suitable for twospotted spider mite feeding and development. This pest can be found in a variety of indoor plant production situations. If twospotted spider mite is observed on any indoor plants, those plants should be removed from close proximity to other plants. Moving infested plants at least 10 feet or more away from clean plants would be a good start. Promptly addressing a few problem plants is far easier than attempting to control a widespread infestation, even if some plants must be destroyed.

Biological control of twospotted spider mite can be an extremely effective form of management indoors. In greenhouse trials from North Carolina State University, the predatory mite *Phytoseiulus persimilis* was very effective at managing twospotted spider mite. This mite can be purchased from any commercial biological control company. Other biological control agents include minute pirate bugs, *Orius* spp., and lacewing larvae, *Chrysopa* spp. (described in the next section). For chemical control, insecticidal soaps or oils should be considered.

References

Cranshaw, W., Melissa Schreiner, Kadie Britt, Thomas P Kuhar, John McPartland, and Jerome Grant. 2019. Developing insect pest management systems for hemp in the United States: A work in progress. Journal of Integrated Pest Management, Volume 10, Issue 1, 2019, 26, https://doi.org/10.1093/jipm/pmz023

Natural enemy/beneficial insects and mites

Many beneficial insects and mites are present in outdoor hemp fields. It is important that these species are conserved. If pesticides must be used to manage pest presence, try to use products that will not be harmful to beneficial insects and mites. Below is a list of beneficial insects and mites that can be commonly seen helping to manage pests in hemp.

- Lady beetle adults and larvae. Convergent lady beetle, *Hippodamia convergens*, multicolored Asian lady beetle, *Harmonia axyridis*, and spotted lady beetle, *Coleomegilla maculata*
- Green lacewing adults and larvae, *Chrysopa spp.* or *Chrysoperla spp.*
- Minute pirate bug adults and nymphs, *Orius spp.*
- Damsel bug adults and nymphs, *Nabis spp.*
- Big-eyed bug adults and nymphs, *Geocoris spp.*
- Predatory stink bug adults and nymphs, *Podisus spp.* and *Euthyrhynchus floridanus*
- Spiders
- Parasitoid wasps for aphids – at this time, we are not sure of particular species.
    - Many times, aphid ‘mummies’ can be found on hemp leaves (see photo below). These are shiny tan/brown in color and look like bloated aphids. Mummies are excellent confirmation that aphids that have been parasitized by a very small, predatory wasp. Parasitic wasps lay eggs in aphid bodies. Wasp eggs use the aphid body as a source of food and site for development. The aphid will be killed by this and a parasitic wasp will hatch out.
    - Parasitoid wasps are very small (gnat-sized) and will not harm humans.

- Predatory mites for twospotted spider mite, *Phytoseiulus persimilis* or *Amblyseius fallicus*

Insect and mite classification

The most concerning insect and mite pests for hemp production have been outlined. However, many other species can be found in hemp. This insect and mite classification list has been compiled based upon field and indoor observations of hemp grown in Virginia. Sporadic or fleeting infestations of insects and mites can and will occur. Populations of these pests may reach levels where feeding can be injurious to hemp, but their presence is not as consistent as the major pests outlined above. Below, the insects and mites observed in hemp are classified by host plant preference (generalist or specialist), type of mouthparts (chewing or piercing-sucking), and area of plant impacted by feeding (leaf/foliar, stem/stalk, flower bud/seed, or root).

	Host plant preference		Type of mouthparts		
Pest	**Generalist**	**Specialist**	**Chewing**	**Piercing-sucking**	**Borer**
Corn earworm	✓		✓		
Hemp russet mite		✓		✓	
Cannabis aphid		✓		✓	
Twospotted spider mite	✓			✓	
Armyworms	✓		✓		
Cucumber beetle	✓		✓		
Japanese beetle	✓		✓		
Grasshoppers	✓		✓		
Leafhoppers	✓			✓	
Tarnished plant bug	✓			✓	
European corn borer	✓		✓		✓
Stink bugs	✓			✓	
Rice root aphid	✓			✓	
Termites	✓		✓		
Fire ants	✓		✓		
Wireworms	✓		✓		
Other caterpillars	✓		✓		

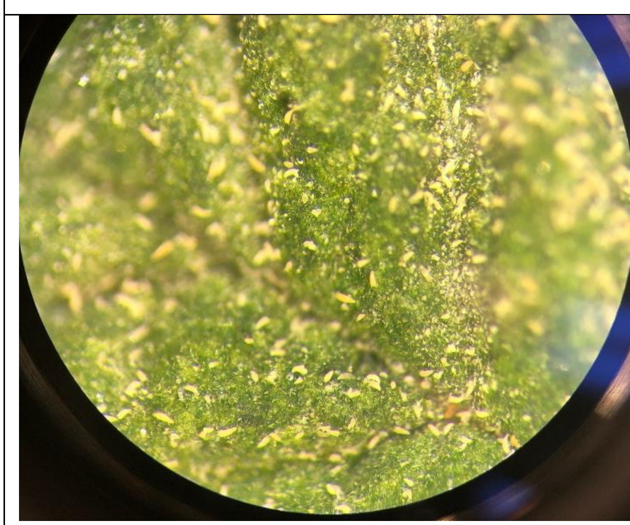
	Area of plant impacted by feeding			
Pest	Leaf/foliar	Stem/stalk	Flower bud/seed	Root
Corn earworm			✓	
Hemp russet mite	✓		✓	
Cannabis aphid	✓	✓	✓	
Twospotted spider mite	✓			
Armyworms	✓			
Cucumber beetle	✓			
Japanese beetle	✓			
Grasshoppers	✓	✓		
Leafhoppers	✓			
Tarnished plant bug	✓		✓	
European corn borer		✓	✓	
Stink bugs			✓	
Rice root aphid				✓
Termites		✓		✓
Fire ants		✓		✓
Wireworms				✓
Other caterpillars	✓			

Color photo 1: bud rot in floral hemp bud from corn earworm feeding.

Color photo 2: adult corn earworm moth on hemp plant.

Color photo 3: young corn earworm larva.

Color photo 4: later stage corn earworm larvae.



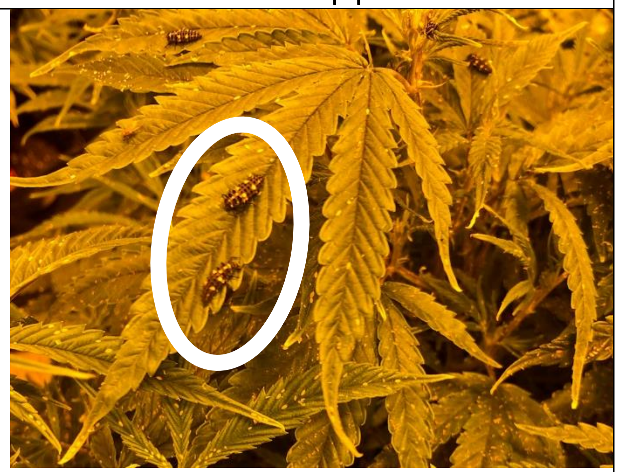
Color photo 5: hemp russet mite population seen on hemp leaf under microscopy.

Color photo 6: upward curling of hemp leaves as a result of hemp russet mite feeding.

Color photo 7: bronzing/russeting of hemp leaf tissue as a result of hemp russet mite feeding.

Color photo 8: cannabis aphid infestation on indoor hemp plant.

Color photo 9: cannabis aphid skins caught in honeydew on surface of hemp leaves.



Color photo 10: lady beetle larvae consuming cannabis aphids (seen inside white oval).

Color photo 11: twospotted spider mite feeding injury – stippling on hemp leaf.

Color photo 12: twospotted spider mite seen under microscopy.