Contribution of different host plants to the adult population of western bean cutworm

Dakota C. Bunn[1], Eduardo Dias de Oliveira[2], Frederick Springborn[3] Miquel A. Gonzalez-Meler[2] and Nicholas Miller[1]

1. Department of Biological Sciences, Illinois Institute of Technology, Chicago, IL 60616
2. Ecology and Evolution, Department of Biological Sciences, University of Illinois at Chicago, 845 West Taylor St, Chicago IL-60527
3. Michigan State University Extension, Montcalm County, Stanton, MI 48888

Abstract

The western bean cutworm, *Striacosta albicosta* (Smith) (Lepidoptera: Noctuidae), is historically a pest of both corn (*Zea maize* L.) and dry beans in the western Great Plains. However, it has recently undergone an eastward range expansion establishing itself across the corn belt in twenty-five states and four Canadian provinces. To mitigate the effects of infestation in Michigan, foliar insecticides are used in beans whereas management of the pest in corn relies more heavily on the use of Bt-expressing hybrids. In this study stable carbon isotope analysis was used to determine what crop adult moths developed on as larvae with analysis showing that very few of the adult moths developed on beans in their larval state. These results suggest that beans and corn are not suitable as co-refuges and that mainly adults which developed on corn are contributing to the next generation of western bean cutworm in Michigan.

Introduction

The western bean cutworm, *Striacosta albicosta* Smith (Lepidoptera: Noctuidae), is a univoltine pest of both corn (*Zea maize* L.) and dry beans (*Phaseolus* sp L.). In corn, direct feeding with an infestation of one larva per ear can result in a loss of four bushels per acre, with large infestations reducing the yield by 30 to 40%, while in beans the yield is reduced by 8 to 10% (Peairs 2014). Additionally, direct feeding provides entry for molds and other plant pathogens, increasing the risk of diseases and mycotoxin contamination (Rice and Dorhout 2006, Seymour et al. 2010, Difonzo et al. 2015). Historically a pest of the western Great Plains region of the United States, over the past quarter of a century it has expanded eastward and is now established in at least twenty-five states and four Canadian provinces (Hoerner 1948, Hagen 1962, Jeschke 2018). In Michigan, the western bean cutworm was first detected in 2006 and severe injury to field corn was observed the following season (Smith et al. 2018).

The western bean cutworm prepupae overwinter before pupating in the spring. Adults then emerge and lay eggs from the end of June to mid-August (Blickenstaff 1979). In corn, oviposition occurs near the whorls on the upper side of the leaf just before tasseling. Once hatched, the larvae will feed on the tassels before migrating into the ear and feeding on the silks and kernels for the remainder of their development (Hagen 1962). In dry beans, eggs are laid on the underside of the leaf and once hatched the larvae feed nocturnally on the leaves and buds of the plants. Once further developed, the larvae feed on the developing seeds and pods at night while moving to the soil during the day. On both plants, once the larvae reach the sixth instar, they then burrow into the soil and create overwintering chambers before the process starts anew (Hoerner 1948, Seymour et al. 2010).

Despite the fact that the western bean cutworm feeds on two very different plants it is not a generalist herbivore. Larvae can successfully develop on several species of *Phaseolus* and some related legume species. However, they are unable to develop on soybeans (*Glycine max*). Nor are larvae able to develop on tomatoes (*Solanum lycopersicum*) or the ancestor of domesticated corn, teosinte (*Zea mays* spp *mexicana*) (Blickenstaff and Jolley 1982).

Management of the western bean cutworm varies depending on the host and time of the growing season. In beans, foliar insecticide applications are the main mechanism of control, with pyrethroid applications showing excellent control of the western bean cutworm (Difonzo et al. 2015). However, application of foliar insecticides requires labor intensive scouting practices which are challenging due to the location of the egg masses and the nocturnal habits of the larvae. Another scouting method requires the use of pheromone traps to capture adult moths to determine when insecticide treatment is needed. Unfortunately, established insecticide application thresholds have been unreliable in the Great Lakes region where significant bean and corn damage is still observed when trap numbers are low, compared to native western regions such as the state of Nebraska (Difonzo et al. 2015). In corn, foliar insecticide treatments are occasionally used, but they are ineffective once the larvae have entered the ear (Smith et al. 2019). Due to this, transgenic corn hybrids are a widely used approach to western bean cutworm management.

Transgenic corn hybrids expressing proteins derived from *Bacillus thuringiensis* are used to manage western bean cutworm infestation. Previous studies have demonstrated that Cry1Aa, Cry1Ab, Cry1Ac, and Cry9C proteins are unable to control WBC and that while Cry1F was once effective, WBC has gained tolerance to the toxin (Dyer et al. 2013, Ostrem et al. 2016, Wolff 2018, Smith et al. 2019). Therefore, the only current hybrids that are marketed for successful control of WBC are the ones expressing Vip3A (DiFonzo et al. 2018, Farhan et al. 2018, Smith et al. 2018, 2019). Varying management strategies for the two crop types grown in near proximity of each other highlights a need for further development of insect resistance management (IRM) programs to mitigate the development of insecticide resistance. This subsequently requires further understanding of WBC biology and interactions between adult moths that developed on differing host crops.

Currently IRM programs recommend the use of refuges in corn in order to sustain a population of susceptible target pests (Bates et al. 2005, Onstad et al. 2018, Anderson et al. 2019). The extremely low number of pests that survive exposure to the toxins and/or pesticide are then expected to mate with the much higher number of susceptible pests that were reared on the refuge crop. However, due to the different management strategies employed between different crops, it is important to know how they may affect each other in areas where both crops (dry beans and corn) are grown in close proximity. It is therefore necessary to understand how adult moths that developed on beans are interacting with adult moths that developed on corn to evaluate the potential for synergistic IRM. Should they be interacting and breeding, it may be possible to use dry beans and corn as mutual refuges to help reduce the development of resistance in areas where both crops are grown in proximity.

We hypothesized that adults that developed on corn or beans would fly at the same time of season, interact and subsequently contribute adult moths to the breeding pool for the next generation. To test this hypothesis, it was first necessary to capture adult moths from an area where both beans and corn are grown, followed by determining the host plant used for development in the larval state via stable carbon isotope analysis. This is possible due to the strong isotopic differences of the stable isotopes of carbon ($\delta^{13}C$) found between C3 and C4 plants (Adams et al. 2016, Hambäck et al. 2016, Hobson et al. 2018). Tissues lacking large amounts of cellular turnover, e.g. wings or heads, can be used to determine if the insect developed on C3 (dry beans) or C4 (corn) photosynthesizing plants.

Material and methods

Sample Collection

Sample collection was performed in central Michigan throughout adult western bean cutworm flight season (mid-July through mid-August) during 2017 and 2018. Nine to twelve traps were used at three to six sites (Figure 1). At each site, traps were placed in between adjacent fields of corn and beans that had been rotated from the other crop the previous year. Trap sites were spaced between 2.8 and 31.4 km with individual traps at each site placed approximately 150 to 1,000 m apart to ensure that they were operating independently.

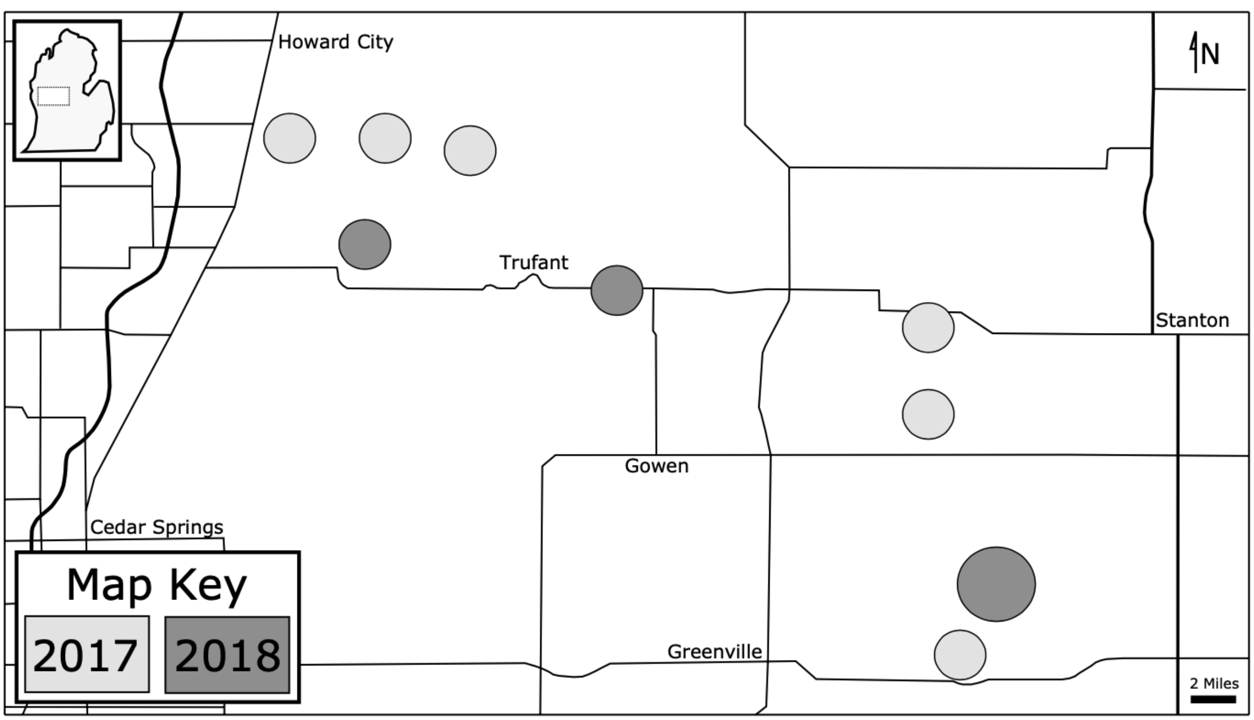

Figure 1. Map of capture sites in 2017 and 2018.

In 2017 and 2018, we used six “bucket” type universal moth traps (Great Lakes IPM Inc., Vestaburg, Michigan), that were baited with western bean cutworm sex pheromone lures (Scentry Biologicals Inc., Billings, Montana). In 2017, we unsuccessfully deployed six small portable LED blacklight traps (White et al. 2016) in an attempt to capture male and female moths. Therefore in 2018, three Quantum Black Light Traps (Leptraps LLC, Georgetown, Kentucky) were used in lieu of the portable LED blacklight traps. All traps contained a piece of insecticidal strip (Herocon Vaportape) to prevent moths from escaping or becoming damaged due to prolonged activity in the enclosed space. Traps were checked every morning and moths were placed in 50 mL centrifuge tubes containing 95% ethanol, returned to the lab and stored at -80°C until they were processed for isotope analysis.

Stable Carbon Isotope Analysis

Samples were selected from periods of peak flight and given unique identifiers. Wings have historically been used for stable isotope analysis in moths due to slow carbon turnover (Adams et al. 2016). For this study we examined both wings and heads. Wings and heads were dried in an oven at 50°C until dry weight was constant (typically 48 hours) to evaporate any traces of ethanol and moisture. Dried wings, generally weighing between 1 to 4 mg, were ground and packaged using forceps into tin capsules for elemental and stable isotope analyses. Heads were packaged whole once dried.

Mass spectrometry was performed via standard protocols as described by Gonzalez-Meler et al. (2017). In short, samples were fully combusted to CO_2 using a Costech (Valencia, CA) elemental analyzer equipped with a zero blank autosampler. Stable carbon isotope ratios were analyzed by an isotope ratio mass spectrometer operating in continuous flow (ThermoFinnigan Delta Plus XL equipped with Conflo III) connected to the elemental analyzer. Host plant carbon isotopic ratios were calibrated after host plant material was collected from the sample sites, dried at 60°C until constant dry weight, ground using tungsten microcontainers and a spex mill shaker, weighed into tin cups and analyzed as previously described. The stable carbon isotopic composition was expressed as a delta notation according to:

$$\delta^{13}C = (R_{sample}/R_{std}-1) \times 1000$$

Where R is the ratio of $^{13}C/^{12}C$ of the sample and standard (std). Lab standards were calibrated against NIST standards using the Pee Bee Dolomite scale. Isotope analyses were performed at the University of Illinois at Chicago (USA) with a precision of 0.2 per mille (‰). The $\delta^{13}C$ values from the plants, wings, and heads were then analyzed to determine individual larval hosts of adult moths.

Results

A total of 3,264 adult moths were captured across the two years of trapping (Figures 2 and 3). In 2017, the portable LED light traps were successful in capturing multiple insect species, including other species of Lepidoptera. However, only 2 western bean cutworm moths were obtained whereas the pheromone traps captured 683 moths. In 2018, the Quantum blacklight traps captured 1,808 moths, and the pheromone traps captured 771 moths. In both 2017 and 2018, two peak periods of flight were observed with a clear peak occurring in mid July and a second peak occurring in late July/ early August. Additionally, in 2017, through the use of pheromone traps, only males were obtained and in 2018, through the use of blacklight and pheromone traps, there were 2,202 males and 306 females collected with 71 being too damaged for sex to be determined. Overall 79% of all moths captured in 2018 were male, 17% were female and 4% were undetermined.

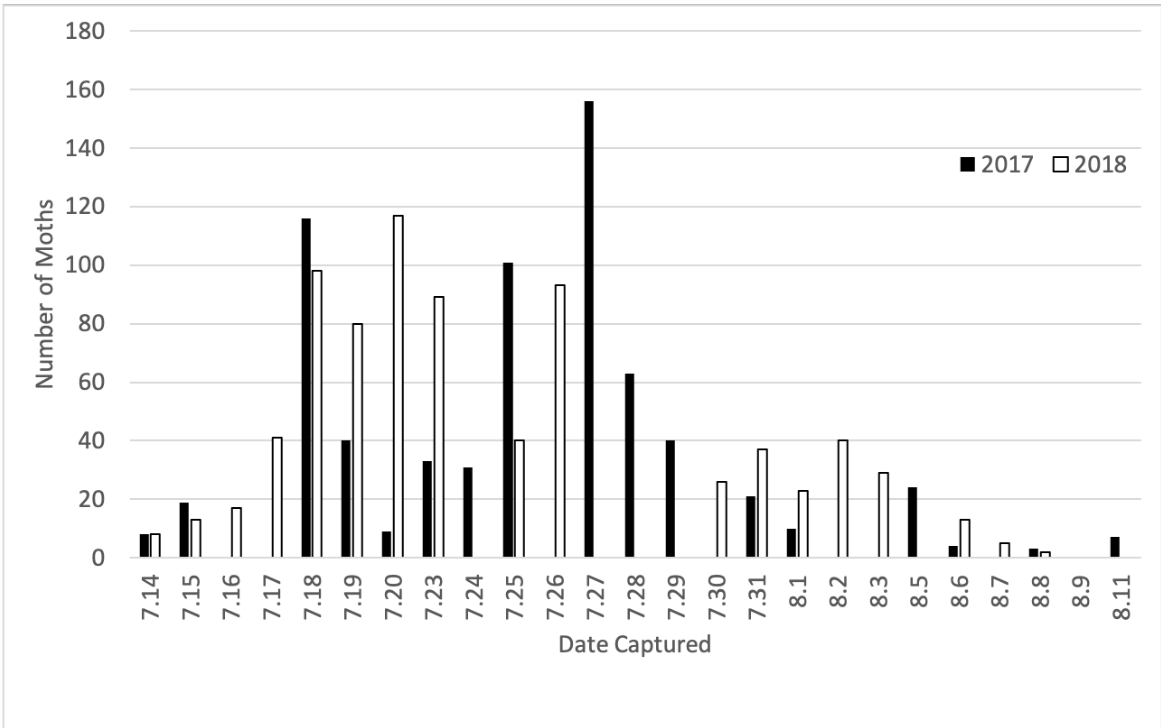

Figure 2. Number of western bean cutworms collected with pheromone traps in 2017 (black) and 2018 (white).

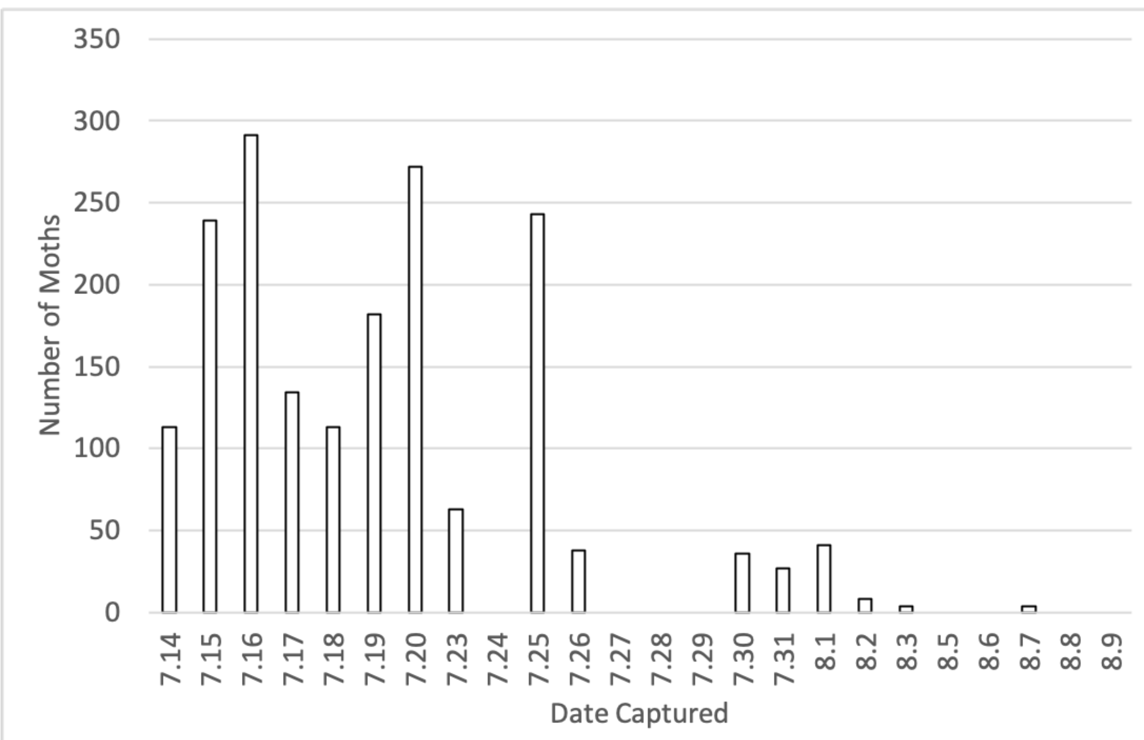

Figure 3. Number of western bean cutworms collected with Quantum blacklight traps in 2018.

Ten percent of the captured moths were used for stable carbon isotope analysis. A total of twenty-four moths were used to determine the effectiveness of head tissue compared to the more traditional wing tissue (Adams et al. 2016). Deviation between wings and heads for each sample was between 0.1 and 2.8 ‰ as shown in Figure 4. Overall, of the 349 moths that were tested, 346 (99.14%) of the moths presented a C4-like isotopic signature with a range of -11.0 to -15.1 ‰, whereas 3 (0.86%) of the moths presented a C3-like isotopic value with a range of -22.3 to -29.4 ‰.

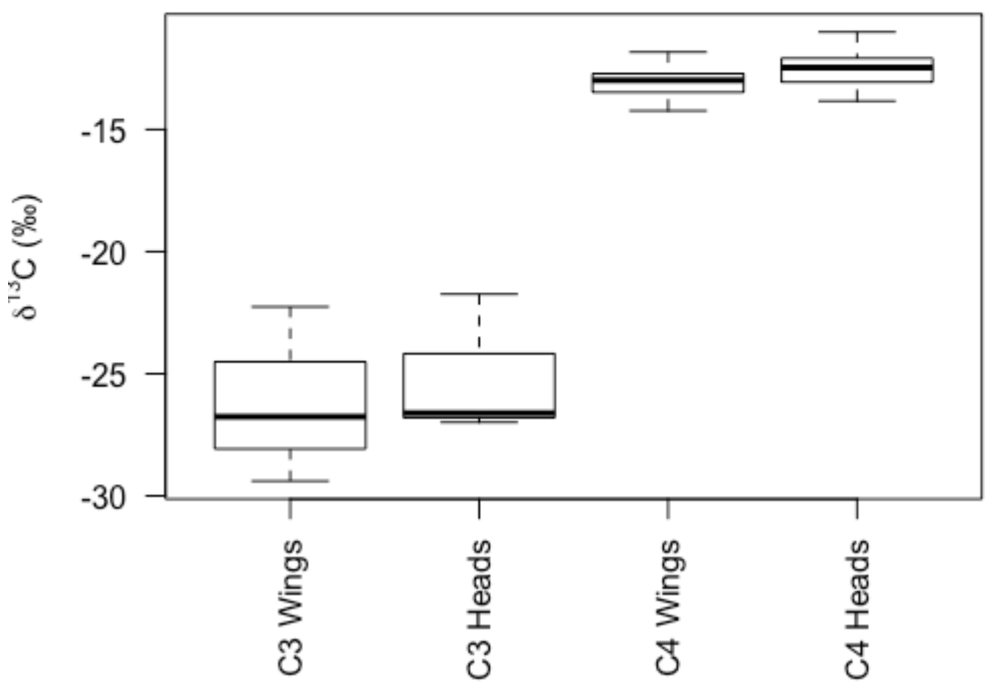

Figure 4. Depiction of the difference between $\delta^{13}C$ (‰) values of heads and wings.

Discussion

Very few moths were found to have developed on beans as larvae, and we found evidence that the western bean cutworms trapped as adults in bean fields developed on corn in their larval state as supported by the stable carbon isotope analysis of adult moths which mimic isotopic values of C4 plants and not C3 plants like beans. Our results suggest that, at least in Michigan, larvae have a low success rate overwintering under bean cultivation. Our results also suggest that corn fields act as an efficient supply of adult moths for the western bean cutworm, maintaining successful mating populations in the region and perpetuating the damage on dry beans the subsequent growing season. It is unlikely that moths that developed on different host plants are flying at a separate time during the season as consistent monitoring performed by extension biologists in the area shows no evidence of differences in developmental timing (Tollini 2018, Springborn 2019). Our results are consistent with laboratory studies, which showed decreased survival of bean feeding larvae before the pupal stage of development when compared to larvae fed on corn (Montezano et al. 2019). Combined with data presented in this study, these results suggest that the environmental factors experienced by the recently established western bean cutworm in Michigan may result in them being unable to fully develop on dry beans, in which case, dry beans may be acting as a regional sink host for this pest.

The portable LED light traps (White et al. 2016) captured only two samples and were unable to be used for the collection of western bean cutworm throughout the 2017 flight season. Subsequently, in 2018 commercial light traps were used, and proved more effective at trapping western bean cutworm moths. These results suggest that large scale trapping efforts for the western bean cutworm should avoid the use of portable LED traps.

In 2018, moths captured in light traps showed a large number of males compared to females. This could suggest an uneven sex ratio among the adult population of western bean cutworm. However, it may also be possible that female western bean cutworms are not flying as frequently and therefore a lower proportion of them were captured in light traps.

Stable carbon isotope analysis of the captured samples showed that both wings and heads can be used to distinguish between moths that developed on beans compared to moths that developed on corn. When performing the study, it was observed that wings were more difficult to package for analysis due to issues caused by poor integrity of the wing tissue and subsequent loss of sample material once dried and ground. This resulted in a newly trained scientist requiring eight hours to package twenty samples. When heads were used, however, they required decidedly less handling time, where twenty samples only required two hours to package. This allowed for a larger number of samples to be analyzed with less hands-on time required and increased productivity of the analysis by 75%.

While there was an increased variance between the $\delta^{13}C$ values of moths that developed on C3 plants compared to moths that developed on C4 plants, the values are within the expected range. Unlike C4 plants that perform photosynthesis across two different cell types to conserve water, C3 photosynthesis takes place in a single cell and is prone to various levels of gas exchange through the stomata depending on humidity and temperature. Subsequently, the variance of the $\delta^{13}C$ in moths that developed on C3 plants is likely due to the relationship between the stomatal conductance and the individual plants photosynthetic rate (Farquhar et al. 1989). Overall, these results would suggest that the process of stable carbon isotope analysis for lepidoptera could be improved through the use of head tissue, eliminating the issues caused by poor integrity of the wing tissue and subsequent loss of sample material, providing a more effective and efficient way to perform population structure and migration studies and sampling in the future. However, further studies with different species are required to ensure that this is not a trait that is unique to the western bean cutworm.

Our study suggests that environmental factors experienced in central Michigan result in moths that developed on corn as larvae being the primary contributors to the mating pool and subsequently the next generation of western bean cutworm in this region. While currently undetermined, these environmental factors may include insecticide application and crop management, soil type and nutrition, as well as travel and migratory patterns of the adult moths. Our results demonstrate that beans and corn cannot be used as mutual refuge in IPM, at least in central Michigan and that further research is needed to determine proper IPM for areas where corn and dry beans are grown in close proximity.

References

Adams, M.-O., C. L. Seifert, L. Lehner, C. Truxa, W. Wanek, and K. Fiedler. **2016**. Stable isotope signatures reflect dietary diversity in European forest moths. Frontiers in Zoology. 13.

Anderson, J. A., P. C. Ellsworth, J. C. Faria, G. P. Head, M. D. K. Owen, C. D. Pilcher, A. M. Shelton, and M. Meissle. **2019**. Genetically engineered crops: importance of diversified integrated pest management for agricultural sustainability. Frontiers in Bioengineering and Biotechnology. 7.

Bates, S. L., J.-Z. Zhao, R. T. Roush, and A. M. Shelton. **2005**. Insect resistance management in GM crops: past, present and future. Nature Biotechnology. 23: 57–62.

Blickenstaff, C. C. 1979. History and biology of the western bean cutworm in Southern Idaho. 1942-1977. Bulletin No. 592.

Blickenstaff, C. C., and P. M. Jolley. **1982**. Host plants of western bean cutworm. Environmental Entomology. 11: 421–425.

Difonzo, C. D., M. M. Chludzinski, M. R. Jewett, and F. Springborn. **2015**. Impact of western bean cutworm (Lepidoptera: Noctuidae) infestation and insecticide treatments on damage and marketable yield of Michigan dry beans. Journal of Economic Entomology. 108: 583–591.

DiFonzo, C., P. Porter, and K. Tilmon. **2018**. Bt trait table January 2018. (https://lubbock.tamu.edu/files/2018/01/BtTraitTableJan2018.pdf).

Dyer, J. M., T. W. Sappington, and B. S. Coates. **2013**. Evaluation of tolerance to *Bacillus thuringiensis* toxins among laboratory-reared western bean cutworm (Lepidoptera: Noctuidae). Journal of Economic Entomology. 106: 2467–2472.

Farhan, Y., J. L. Smith, and A. W. Schaafsma. **2018**. Baseline susceptibility of *Striacosta albicosta* (Lepidoptera: Noctuidae) in Ontario, Canada to Vip3A *Bacillus thuringiensis* protein. Journal of Economic Entomology; Lanham. 111: 65–71.

Farquhar, G. D., J. R. Ehleringer, and K. T. Hubick. **1989**. Carbon isotope discrimination and photosynthesis. 40: 503–537.

Gonzalez-Meler, M. A., L. B. C. Silva, E. Dias-De-Oliveira, C. E. Flower, and C. A. Martinez. **2017**. Experimental air warming of a *Stylosanthes capitata*, vogel dominated tropical pasture affects soil respiration and nitrogen dynamics. Frontiers in Plant Science. 8.

Hagen, A. F. 1962. The biology and control of the western bean cutworm in dent corn in Nebraska. Journal of Economic Entomology. 55: 628–631.

Hambäck, P. A., E. Weingartner, L. Dalén, H. Wirta, and T. Roslin. **2016**. Spatial subsidies in spider diets vary with shoreline structure: complementary evidence from molecular diet analysis and stable isotopes. Ecology and Evolution. 6: 8431–8439.

Hobson, K. A., K. Doward, K. J. Kardynal, and J. N. Mcneil. **2018**. Inferring origins of migrating insects using isoscapes: a case study using the true armyworm, *Mythimna unipuncta*, in North America. Ecological Entomology. 43: 332–341.

Hoerner, J. L. 1948. The cutworm *Loxagrotis albicosta* on beans. Journal of Economic Entomology. 41: 631–635.

Jeschke, M. 2018. Western bean cutworm, corn. DuPont Pioneer. (https://www.pioneer.com/home/site/us/agronomy/crop-management/corn-insect-disease/western-bean-cutworm/).

Montezano, D. G., T. E. Hunt, A. Specht, P. M. C. Luz, and J. A. Peterson. **2019**. Life-history parameters of *Striacosta albicosta* (Lepidoptera: Noctuidae) under laboratory conditions. Journal of Insect Science. 19.

Onstad, D. W., A. L. B. Crespo, Z. Pan, P. R. Crain, S. D. Thompson, C. D. Pilcher, and A. Sethi. **2018**. Blended refuge and insect resistance management for insecticidal corn. Environmental Entomology. 47: 210–219.

Ostrem, J. S., Z. Pan, J. L. Flexner, E. Owens, R. Binning, and L. S. Higgins. **2016**. Monitoring susceptibility of western bean cutworm (Lepidoptera: Noctuidae) field populations to *Bacillus thuringiensis* Cry1F protein. Journal of Economic Entomology. 109: 847–853.

Peairs, F. 2014. Western bean cutworm: characteristics and management in corn and dry beans - 5.538. Extension. (https://extension.colostate.edu/topic-areas/insects/western-bean-cutworm-characteristics-and-management-in-corn-and-dry-beans-5-538/).

Rice, M. E., and D. L. Dorhout. **2006**. Western bean cutworm in Iowa, Illinois, Indiana and now Ohio: did biotech corn influence the spread of this pest? *In* Proceedings of the Integrated Crop Management Conference. Presented at the Proceedings of the 16th Annual Integrated Crop Management Conference, Iowa State University, Digital Press.

Seymour, R., G. Hein, and R. Wright. **2010**. Western bean cutworm in corn and dry beans (No. G2013), NebGuide. University of Nebraska-Lincoln, Lincoln, Nebraska.

Smith, J. L., T. S. Baute, M. M. Sebright, A. W. Schaafsma, and C. D. DiFonzo. **2018**. Establishment of *Striacosta albicosta* (Lepidoptera: Noctuidae) as a primary pest of corn in the Great Lakes region. Journal of Economic Entomology; Lanham. 111: 1732–1744.

Smith, J. L., C. D. Difonzo, T. S. Baute, A. P. Michel, and C. H. Krupke. **2019**. Ecology and management of the western bean cutworm (Lepidoptera: Noctuidae) in corn and dry beans—Revision with focus on the Great Lakes region. Journal of Integrated Pest Management. 10.

Springborn, F. 2019. Western bean cutworm is alive and well in central Michigan. Field Crops. (https://www.canr.msu.edu/news/western-bean-cutworm-is-alive-and-well-in-central-michigan).

Tollini, C. 2018. Western bean cutworm nears peak flight in northeast Michigan. Field Crops. (https://www.canr.msu.edu/news/western-bean-cutworm-nears-peak-flight-in-northeast-michigan).

White, P. J. T., K. Glover, J. Stewart, and A. Rice. **2016**. The technical and performance characteristics of a low-cost, simply constructed, black light moth trap. Journal of Insect

Science. 16.

Wolff, S. 2018. Fighting western bean cutworm and protecting no-till corn yields. (https://www.no-tillfarmer.com/articles/8338-fighting-western-bean-cutworm-and-protecting-no-till-corn-yields?v=preview).