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In This Issue:

Yes it's true: Genetically engineered mosquitoes

Modifying the genes of mosquitoes can transform these pests into a weapon capable of fighting disease. Zika, dengue and chikungunya are all viral diseases spread by the yellow fever mosquito, *Aedes aegypti*. Because there are no cures for these viruses, health officials are aeager to find new methods to slow their spread. Genetically engineered mosquitoes are being studied as a tool for this reason (Fig. 1).

Figure 1. Aedes aegypti mosquito. (Photo credit: https://commons.wikimedia.org/wiki/File:Aedes_aegypti_feeding.jpg)

Gene modification

Scientists are modifying the genome of *A. aegypti* mosquitoes by inserting a gene into the insect that prevents its offspring from surviving to adulthood. This gene, tTAV (tetracycline repressible transactivator variant), makes a non-toxic protein that essentially ties up the machinery inside the insect's cells, so that genes key to survival are not expressed and the insect dies.

The process works by inserting the tTAV gene variant into male mosquitos, which do not bite or transmit disease. In the rearing facilities where these insects are kept, an antidote of tetracycline (an antibiotic) is given to the mosquitoes to turn off the tTAV gene and prevent it from producing the protein responsible for eventually killing the insect. In the presence of tetracycline, the insects survive and reproduce in the rearing facility; however, when the males are released into the wild, their offspring cannot access the antibiotic and they die before becoming adults. The idea is to release large numbers of these male mosquitoes repeatedly over time, so they mate with wild females and reduce the wild population. As populations of these mosquitoes decline, so does the threat of disease.

Field trials

Oxitec, the biotechnology company responsible for creating these genetically engineered mosquitoes, has received regulatory approvals for open field trials. So far trials have taken place in the Cayman Islands, Panama, Brazil and Malaysia and projects are currently ongoing in Brazil and Grand Cayman. Field releases of the genetically engineered mosquitoes have reduced populations of *A. aegypti* mosquitoes by up to 90 percent at test sites over a period of six months.

In 2016, a trial was planned for the Florida Keys, but it was delayed several times and never happened after strong voter opposition. Plans for a future release of the mosquitoes in the United States are currently awaiting approval from the U.S. Environmental Protection Agency.

Bats: A biological pest control service

Bats are a highly specialized group of mammals that are often misunderstood. There are over 1,300 bat species worldwide and at least 12 species in West Virginia. West Virginia bats range in size from the eastern small-footed myotis (4.5 grams) to the hoary bat (27.5 grams), with the average size of a bat in West Virginia being about 10 grams (Fig. 2).

All 12 species of bats found in West Virginia feed on insects. They are voracious eaters, consuming 50 to 75 percent of their body weight in insects each night.

A primary predator of nocturnal insects, bats can suppress populations of forest and agricultural pests. While one bat consuming between 5 and 7.5 grams of insects a night may not sound like much, consider that there may be 100,000 bats on the local landscape. The amount of insects consumed each night jumps to between 1,100 and 1,650 pounds.

Benefits of bats

Bats provide a significant ecological service by feeding on insects and removing agricultural pests, including those capable of transmitting pathogens to humans from the landscape.

The size of a bat's prey depends on the size of the bat and can range from 1 millimeter (midges and mosquitos) to as large a 50 millimeters (beetles and large moths). Many species of agricultural pests have been found in the diets of bats including June beetles, click beetles, leafhoppers, plant hoppers, spotted cucumber beetles, green stinkbugs and corn earworm moths.

Published estimates of the value of pest suppression from bats averages about $74 per acre; therefore, their value to the U.S. agricultural industry is about $22.9 billion per year.

Dangers of bats

While bats do provide a valuable economic service by consuming insects, they can become a nuisance when they roost in domestic structures.

Like all warm-blooded mammals, bats can carry rabies, and a fungus that causes histoplasmosis in humans can grow in deposits of guano (feces) that can build up beneath bat roosts.

Most commonly bats enter houses through small openings or cracks around the roof and eaves. While bats can be excluded from houses, care must be taken to ensure that no bats are trapped inside as they could find their way into the residence or die and create bad odors. It is also important to time bat removal or exclusion to ensure that non-flying bats (young of the year) are not left inside.

Figure 2. A hoary bat, one of at least 12 species of bats in West Virginia, that feed on insects and other agricultural pests. (Photo credit: www.flickr.com/photos/40928097@N07/27639375038)

Because of these considerations and the disease concerns associated with bats, it is recommended that only trained, immunized professionals work on bat removal and exclusion projects.

If you suspect bats are roosting in your home or outdoor structure, please contact the local West Virginia Division of Natural Resources office or private nuisance wildlife control operator for assistance.

Decline in bat populations

Currently, several bat species are facing significant population declines from an introduced disease called white-nose syndrome.

White-nose syndrome is caused by a fungus and impacts hibernating bats. In some hibernacula, hibernating bats have experienced 90 to 100 percent mortality. It is estimated that we have lost over 5.7 million bats in the eastern U.S. Not only are we losing these iconic species from the landscape, but we are also losing their suppression of our insects.

White-nose syndrome has been documented in West Virginia, and we are seeing the impacts of this disease in our bat populations.

– continued on page 5 –

Biological control of plant diseases gaining momentum

Over the last few decades, indiscriminate use of synthetic pesticides has created concerns due to its detrimental effect on the environment and human health. These concerns, however, generated a continued and increased interest in finding environmentally friendly pest management tools.

A significant global effort is underway to generate low-cost, biorational, natural pest control products. Besides crop plant resistance, various biological control methods based on natural pest suppressing organisms are regarded as main alternatives. More specifically, biological control of plant disease has been gaining momentum in recent years because it offers an alternative and supplement to synthetic chemicals. Microorganisms from diverse groups have successfully been used as biocontrol agents due to their ability to suppress harmful microbes using a variety of mechanisms, such as competition, antibiosis (production of antimicrobial compounds) and resistance induction in the host plant.

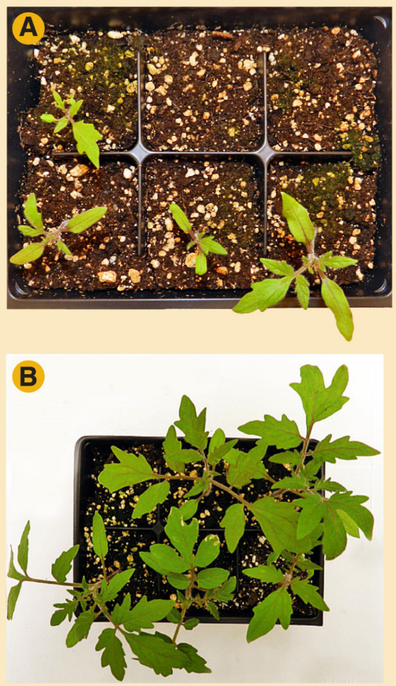

Figure 3. Difference in tomato seedling vigor: a) seed and non-pasteurized planting mix treated with Serenade soil; b) seed and pasteurized planting mix treated with Serenade soil. (Photo credit: MM Rahman)

History of biological control

Since the inception of agriculture, the biological control of plant disease has played a significant role in integrated disease management by balancing harmful and beneficial microbes. Although it took scientists and pathologists a long time to discover and characterize these beneficial microbes, current knowledge and the availability of beneficial microbe-based commercial products has changed growers' perceptions of biological control of plant diseases. As a result, biocontrol is becoming a more regular part of many growers' integrated disease management programs.

As consumers demand more sustainable production methods utilizing biorational products, biological control has found its place in the form of augmentative releases, particularly for the management of pests that are difficult to control with pesticides or are prone to developing resistance against chemicals. Retailers have taken notice and many are beginning to set expectations for what they want to see from growers.

Due to increased investment in research and education on multiple aspects, including new products, application techniques and supply chain considerations, biocontrol has proven a good fit for growers looking to add more sustainable tools in their production.

Considerations

A few applied considerations can help improve efficacy of products at the growers' level.

- Biocontrol agents are living organisms that interact with environmental factors and other living organisms in the application sites; therefore, application of biocontrol products should be done in a cool, moist environment.
- Proactively apply biocontrol agents at the very early stage of disease as opposed to when the disease has already set in.
- Biocontrol agents should be in place to colonize the plant surface before encountering harmful microbes. For example, treat seed and planting mix with biocontrol agents to facilitate root colonization. A recent study showed that pasteurized planting mix treatment with biocontrol agents followed by seeding provided maximum colonization of seedling root systems and enhanced seedling vigor compared with non-pasteurized planting mix treatment (Fig. 3). This application method is especially important for biocontrol agents that do not flourish in a competitive environment.

– continued on page 4 –

Using growing degree days to predict germination

Seed germination occurs when it has completed its dormancy requirements and when the environmental conditions are favorable for subsequent growth of seedlings upon germination. Most weeds that compete with crops during the growing season or interfere with lawns and landscape plantings germinate in spring (summer annuals) as the ambient air temperature and soil temperature start to rise.

Growing degree days

Apart from temperature, adequate soil moisture, oxygen and light are other requirements for seed germination. Typically, these are met under normal circumstances so the factor that primarily determines seed germination is air/soil temperature. Each weed has a biological clock (the seed) that keeps track of warming trends in its immediate microenvironment. Once a cumulative threshold is achieved, it commits to germination as long as other factors are suitable. This innate record-keeping mechanism of a seed, referred to as Growing Degree Days (GDD), can be considered as heat units stored in its memory.

Typically, GDD is calculated as the difference of the mean temperature and base temperature, usually 50 F (referred to as GDD_{50}), for a given day. GDD_{50} accrued from the beginning of Julian calendar (January 1) can be used to predict germination of certain common weeds. For example, smooth crabgrass *(Digitaria ischaemum)* begins to germinate when the GDD_{50} accumulates to about 150; however, large crabgrass *(Digitaria sanguinalis)* requires 150 to 200 more GDD_{50} units to germinate. Certain phenological indicators, such as forsythia bloom drop, are used to estimate GDD accumulation that coincide with smooth crabgrass germination.

Determining GDD

Not all populations of weeds belonging to the same species germinate at the same time; a small percentage may germinate early and be followed by a peak germination period. Some species have a narrow window of GDD requirements, while others have a broad range.

Figure 4. The invasive weed joint-head Arthraxon (Arthraxon hispidus) emerges when GDD_{50} approaches 175; photo taken when GDD_{50} recorded 217. (Photo credit: R. Chandran)

Online tools are available to determine GDD for a given location. A tool on the Climate Smart Farming website at Cornell University *(http://climatesmartfarming.org/tools/csf-growing-degree-day-calculator/)* determines GDD for a given location once the address of the location is entered. By keeping track of GDD using this tool, one can maintain a record of weed germination in the backyard, especially during early- to mid-spring for future reference.

The GDD model can be used to time a management method to control a given pest if its GDD requirements are known. For example, the application of mulch or a pre-emergent herbicide can be timed when the GDD_{50} approaches 150 to manage smooth crabgrass in lawns. Similarly, the invasive weed joint-head Arthraxon *(Arthraxon hispidus)* emerges when GDD_{50} approaches 175 (Fig. 4).

Biological control of plant diseases gaining momentum

– continued from page 3 –

- Application of biocontrol agents on an augmentative basis may need more frequent applications early in the season until a significant amount of the biocontrol agent has built up.
- Producers of agricultural commodities who are willing to integrate biocontrol agents into their crop protection package should look for products listed on the Organic Material Review Institute website *(http://www.omri.org/omri-lists)*. Because efficacy of products has not been verified by university scientists, it is imperative to retrieve efficacy data from university scientists before using a specific product.

Using banker plants in an IPM program

Biological control agents have proven very effective in managing several pests in high tunnels and greenhouses; however, some of the challenges include the cost of purchasing biological control agents and keeping those parasitoids and predators in the structure.

Consider using banker plant systems as part of your biological control plan. A banker plant system supplies a non-pest prey for parasitoids and predators to use as an alternative food source. The plants housing the non-pest prey also provides a home for beneficial insects promoting reproduction; thus, providing the next generation of pest assassins. This relationship reduces the need to purchase fresh natural enemies.

Producing banker plants to help maintain this parasite includes growing cereal grains in a pot (or hanging basket) and introducing cereal aphids to create a population high enough to sustain the parasite population (Fig. 5). The plants will continue to produce more parasitoids, but when the cereal aphid population is low, fresh banker plants will be needed to replace the previous ones.

Figure 5. Producing banker plants. (Photo credit: Chelsie Chapman)

The most common banker plant system is one that uses a non-pest aphid species, cereal aphid (bird cherry oat aphid, *Rhopalosiphum padi*), that lives and feeds exclusively on cereal plants (Fig. 6). This system is often used in greenhouses to help maintain the parasite (*Aphidius colemani*) which is used to manage aphid pest species.

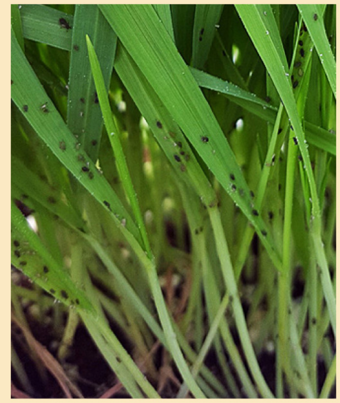

Figure 6. Cereal aphids on banker plants. (Photo credit: Chelsie Chapman)

The recommendation is to start with two banker plants per greenhouse and to add one each week. It is critical to remember to continue producing new banker plants to add throughout the season. Banker plants should be added as soon as plants are introduced. Waiting until you see your first aphid pest is too late, as it can take three to four weeks before new adult parasites will be produced.

West Virginia State University started producing banker plants last spring and found them easy to create. It was also found that with the use of banker plants, our parasitoids and predators were maintained for a longer duration of time. Banker plants will continue to be added this year as part of the IPM program to support and extend purchased biological control agents.

Bats: A biological pest control service

– continued from page 2 –

West Virginians can help combat this population decline by:

- Promoting bat conservation by understanding the ecological services that our bats provide and educating landowners of their importance.
- Avoiding caves or mines where bats may be hibernating during the winter months. If you enter those caves or mines outside of the hibernation period, be sure to decontaminate your clothing or gear before you enter another cave to help stop the spread of the white-nose fungus.
- Constructing bat houses to provide daytime roosting sites.

Food for thought: Corn earworm management

Corn earworm, *Helicoverpa zea*, is the primary economic pest of sweet corn in West Virginia (Fig. 7). Infestation levels in the Northeast region of the United States vary with the year, time of season and farm location. Infestations of corn earworm in West Virginia result from migrant moths carried northward on storm fronts during mid- to late summer. Sweet corn producers rely on timely pest monitoring and insecticide sprays to control this pest. However, insecticide control programs are costly, pose exposure risks to applicators, require considerable time and management, and can negatively impact non-target organisms and the environment.

Insecticide usage

When using insecticides for corn earworm control, it is important to time the first spray at early silking and then apply subsequent sprays on a schedule based on the flight activity of moths. Most corn earworm eggs are laid directly on the silks. Once larvae hatch, they quickly move down the silk channel and begin feeding on the ear tip, where they are protected from insecticide sprays. Therefore, it is necessary to target larvae before they enter the ear by treating silk tissue when moth pressure is high.

Pyrethroid insecticides (beta-cyfluthrin, bifenthrin, cyfluthrin, esfenvalerate, lambda-cyhalothrin, permethrin, zeta-cypermethrin) have been the traditional choice for control, but their control efficacy has declined in certain areas due to resistance in corn earworm populations.

Spray mixtures containing an alternative active ingredient plus a pyrethroid are often used to circumvent resistance development and improve control of other sweet corn pests. Rotations and mixtures with different active ingredients, such as chlorantraniliprole (Coragen), methomyl (Lannate), spinetoram (Radiant) and spinosad (Blackhawk), are increasingly used and can provide control. However, it is widely recognized that pyrethroids no longer provide enough ear protection making it necessary to switch to alternative products.

Bt sweet corn

The problems and challenges with foliar insecticide applications can essentially be eliminated with Bt sweet corn, which expresses insect toxic proteins from the bacterium, *Bacillus thuringiensis* (Bt), in plant tissue. This technology has revolutionized the way many corn insect pests are managed. There are three types of Bt sweet corn commercially available for lepidopteran pests: Attribute® hybrids (expressing Cry1Ab protein) and Attribute® II hybrids (expressing the Cry1Ab and Vip3A proteins) both from Syngenta Seeds, as well as Performance Series™ hybrids (expressing the Cry1A.105 and Cry2Ab2 proteins) from Seminis Seeds.

Figure 7. Corn earworm larva. (Photo credit: Daniel Frank)

Attribute® hybrids have been commercially available since 1998, and acreage of this hybrid has increased significantly since its introduction. However, effectiveness of the Cry1Ab protein and the Attribute® hybrid has varied for controlling corn earworm, particularly in the southern United States. Recent field trials conducted in West Virginia have shown that Attribute® II hybrids can provide excellent control of corn earworm larvae. Growers using Attribute® II hybrids would likely not need to use supplemental insecticidal sprays, except for secondary pests.

Due to the yearly migration of potentially Cry1Ab-resistant moths from southern regions, the risk of further resistance development in the entire Northeast will likely increase and may compromise the reliability and durability of other Bt proteins.

Integrated pest management can further delay the evolution of resistance and must also be incorporated into future corn earworm management plans. IPM practices, such as monitoring, crop rotation, rotation of Bt hybrids that produce different Bt proteins and judicious use of insecticides, are all highly compatible with the goals of IPM and insect resistance management for corn earworm.

Chilling requirements for fruit trees

Through evolution, plants have adapted to the drastic changes of their natural environment allowing them to thrive. Trees during the winter months live in a stage of suspended animation, a form of hibernation referred to as dormancy. Dormancy is their built-in defense mechanism against winter cold injury.

Dormancy

In the fall, plants start entering endodormancy. Endodormancy, or deep dormancy, is where the suspension is regulated by the inner metabolic processes within the meristem, as well as factors that suppress growth within the plant. To get out of endodormancy, plants must be exposed to cold temperatures, between 32 and 50 F, for a certain amount of time. Exposure to cold temperatures is referred to as chill hours.

Figure 8. These trees did not receive adequate chilling hours thereby expressing different developmental phases ranging from bud-break to flowering, green fruit, and mature fruit, simultaneously on the same tree. (Photo credits: M. Danilovich)

Chill hours

Temperatures below freezing increase the winter hardiness of the plants and make them less prone to winter cold injury. The number of required chill hours varies among different plants from about 100 hours in tropical and sub-tropical climates up to 1,500 hours in northern areas. The winters in the southern regions are brief, meaning plants have shorter chill requirements. Plants in the extreme north are low-chill because the winters are long with temperatures below zero and only a few short, spring warm-ups.

Once the chilling requirements are satisfied, the plants enter into endodormancy. After the outside conditions are favorable, the plants will start growing again.

Ecodormancy is imposed by environmental conditions, such as extreme temperatures, drought, etc. As temperatures start getting into the mid-40s, the buds will start to swell, pushing the bud scales to separate. The bud development becomes apparent once the buds become swollen and start to show green. Once budded, plants lose the ability to readjust to colder temperatures, and extreme cold temperatures will cause injury to the buds.

West Virginia trees

In West Virginia, plants have chill requirements ranging from about 400 to 1,500 hours. Most of the fruit species are in the range of 700 to about 1,300 chill hours, though some apples, like Gala and Pink Lady, have a lower chill requirement of 400 hours. At higher elevations, plants have higher chilling requirements to keep them dormant and safe from sudden warm-ups and late frosts. If the plant does not have sufficient exposure to chilling, the buds don't develop and grow. In West Virginia, we normally accumulate enough chill hours by late January to mid-February.

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