

Progress
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April 2014

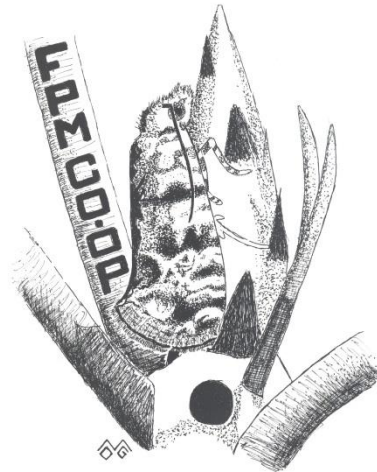
Quarterly Newsletter
on
Forest Pest Management
Issues

PEST is a quarterly newsletter that provides up-to-date information on existing forest pest problems, exotic pests, new pest management technology, and current pesticide registrations in pine seed orchards and plantations. The newsletter focuses on, but is not limited to, issues occurring in the Western Gulf Region (including, Arkansas, Louisiana, Mississippi, Oklahoma, and Texas).



A *Phloeosinus* spp. beetle (pictured above) is actively causing mortality of eastern red cedar around the Round Top, TX region.

Forest Pest Management Cooperative



Nantucket Pine Tip Moth, *Rhyacionia frustrana* (Comstock)

Texas A&M Forest Service, Forest Health,
P.O. Box 310, Lufkin, Texas 75902-0310

**Evaluation of a New Plug Injection
System for Application of PTM™ to
Containerized Seedlings in the Nursery**

By Melissa Fischer

In 2011, a plug injection system was developed by Stewart Boots, S&K Designs to deliver PTM™ (fipronil) to containerized seedlings in the nursery. This method is expected to allow for safer and more efficient application of PTM™ to containerized seedlings for control of pine tip moth.

To test this new method, in 2012, containerized seedlings were individually treated at the nursery prior to planting using the plug injection system. The seedlings were treated with PTM™ at two different rates based on the restricted rate of 59g AI/acre/year and the number of trees planted per acre (TPA). There were three treatments in total:

- Treatment 1: PTMTM: mid-concentration/undiluted plug injection [1.4 mL PTM undiluted/ seedling (435 TPA rate)]
- Treatment 2: PTMTM: low concentration/undiluted plug injection [1.0 mL PTM undiluted/ seedling (600 TPA rate)]
- Treatment 3: control: containerized, untreated

Five recently-harvested tracts were selected across the southeastern United States [TX (Campbell Group), AR (Plum Creek), GA (International Forestry Co.), AL (Rayonier), and NC (Weyerhaeuser)]. Each treatment was randomly selected for placement along a row. Seedlings were planted at 8-foot spacing along each row.

In 2012 and 2013, tip moth damage was evaluated after each tip moth generation by determining the proportion of tips infested on the top whorl. The diameter, height, and volume of all trees were measured in mid- to late November of each year, when tree growth had stopped.

There were no significant differences between treatments 1 and 2, the PTMTM treated seedlings, and therefore the data were analyzed as PTMTM treated containerized seedlings vs. untreated, containerized seedlings.

In 2012, containerized seedlings treated with the plug injection system had significantly less percent tip moth infestations in all tip moth

generations compared with untreated containerized seedlings (Figure 1).

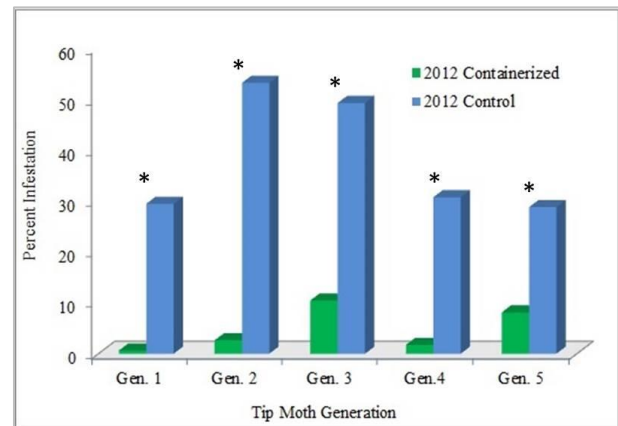


Figure 1. Percent tip moth infestation for containerized seedlings treated with PTMTM using a plug injection system (green bars) and untreated, containerized seedlings (blue bars) in 2012. Asterisks represent generations where there was a significant difference in extent of tip moth infestation between treated and untreated seedlings.

In 2013, containerized seedlings treated with the plug injection system had significantly less percent tip moth infestations in generations 1 and 2 compared with untreated, containerized seedlings, but there were no significant differences in generations 3, 4, or 5 (Figure 2).

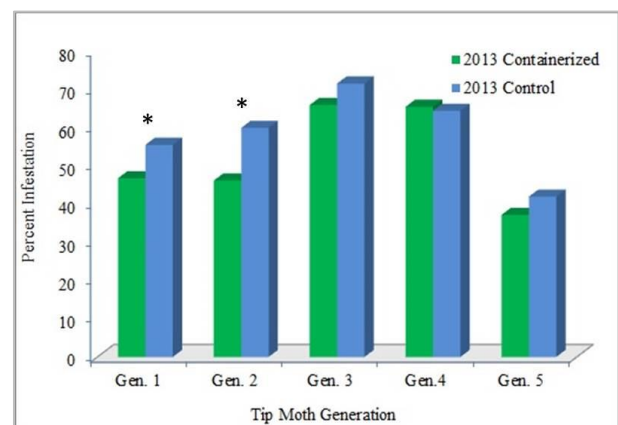


Figure 2. Percent tip moth infestation for containerized seedlings treated with PTMTM using a plug injection system (green bars) and untreated, containerized seedlings (blue bars) in 2013. Asterisks represent generations where there was a significant difference in

extent of tip moth infestation between treated and untreated seedlings.

The diameter, height, and volume of containerized seedlings treated with PTM™ using the plug injection system were significantly greater than those of the containerized, untreated seedlings in 2012 and 2013 (Figures 3, 4, and 5).

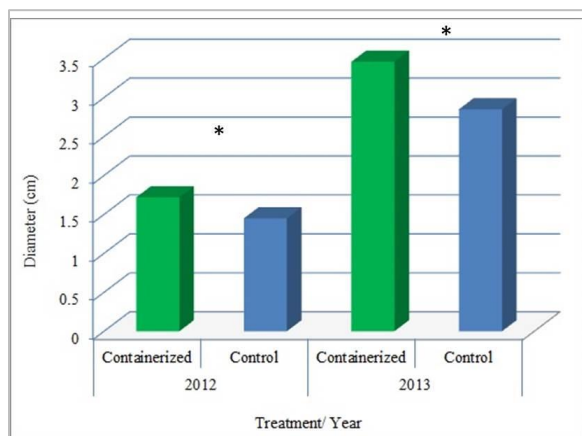


Figure 3. Diameter (cm) of containerized seedlings treated with PTM™ using a plug injection system (green bars) and untreated, containerized seedlings (blue bars) following one year of growth (2012) and two years of growth (2013). Asterisks represent significant differences between containerized, treated seedlings and untreated seedlings.

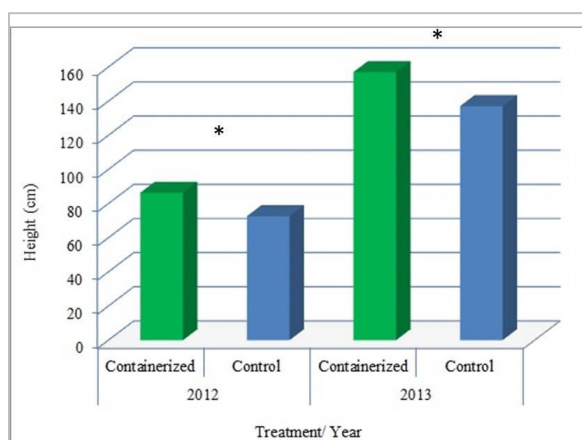


Figure 4. Height (cm) of containerized seedlings treated with PTM™ using a plug injection system (green bars) and untreated, containerized seedlings (blue bars) following one year of growth (2012) and two years of growth (2013). Asterisks represent significant differences between containerized, treated seedlings and untreated seedlings.

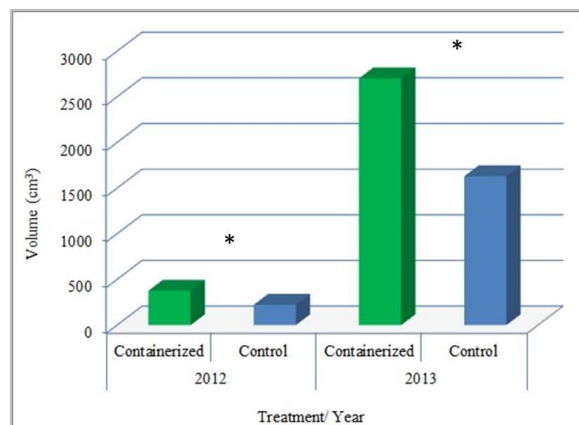


Figure 5. Volume (cm³) of containerized seedlings treated with PTM™ using a plug injection system (green bars) and untreated, containerized seedlings (blue bars) following one year of growth (2012) and two years of growth (2013). Asterisks represent significant differences between containerized, treated seedlings and untreated seedlings.

Based on these results, it is clear that the plug injection system is successful at applying PTM™ to containerized seedlings, resulting in lower pine tip moth infestation and an increase in growth for two years following planting compared to untreated, containerized seedlings.

In the Genome of Loblolly Pine Lies Hope for Better Resistance to a Damaging Disease

March 20, 2014 by zhoyale

U.S. Forest Service Southern Research Station (SRS) scientists co-authored an article published in the journal *Genome Biology* that reports the sequencing, assembly, and annotation of the loblolly pine (*Pinus taeda*) genome. As the primary source of pulpwood and saw timber for the U.S. forest industry, loblolly pine is of great

economic importance to the South and the nation. David Neale, professor of plant sciences at the University of California, Davis, led the loblolly pine genome project.

“The project was a huge undertaking because at 22 gigabases, the loblolly pine genome is about eight times larger than the human genome,” said C. Dana Nelson, SRS Southern Institute for Forest Genetics (SIFG) project leader and research geneticist. “The group chose loblolly pine both because of its economic importance and the knowledge gained from 60 years of breeding the species and managing millions of trees in genetic trials.”

As part of the project, researchers identified a candidate for a gene involved in resistance to fusiform rust, a disease that infects southern pines. SIFG biological science technician Katherine Smith worked with John M. Davis, professor and associate director of the School of Forest Resources and Conservation at the University of Florida (UF), to compare mapped sections of the genome with sections found in loblolly specimens previously inoculated with the pathogen that causes fusiform rust.

“Fusiform rust is the most damaging disease of southern pines—and one of the most complex due to genetic interactions between the pathogen and its host,” said Davis, who also serves as faculty and Executive Committee member at

the UF Genetics Institute. “Genetic resistance is the only realistic way to manage the disease, which infects young trees within their first five years of growth and weakens or girdles the stem. Chemical control is expensive, impractical, and not very good for the environment.”

Researchers and breeders can use the resistance genes as markers to track resistance in pine breeding populations and to guide tree planting at the stand level. “The fusiform rust pathogen has evolved to defeat some rust resistance genes in loblolly,” said Nelson. “The increased molecular understanding from the loblolly genome sequencing effort provides managers with a new effective tool to determine how well seedlings will grow on a particular site.”

SIFG involvement in sequencing the loblolly genome actually goes back at least two decades, when SIFG helped develop an array of resources to help speed up mapping and sequencing the loblolly pine genome and provide the ability to identify genes that influence factors such as tree growth, wood quality, stress tolerance, and resistance to disease. SIFG also conserved and supplied the plant tissue used in the genome sequencing project and provided quality control on the DNA samples that were sequenced.

For more information, contact Dana Nelson at dananelson@fs.fed.us.

The loblolly genome project was led by a team at the University of California, Davis, and the assembly stages were led by Johns Hopkins University and the University of Maryland. Other

collaborating institutions include Indiana University, Bloomington; Texas A&M University; Children's Hospital Oakland Research Institute and Washington State University.

The research was supported in part by the USDA National Institute of Food and Agriculture through its flagship competitive grants program, the Agriculture and Food Research Initiative.

New Parasitoid Wasp Species Found in China May Help Save Pine Trees

February 24, 2014 by Entomology Today

A new wasp species discovered in China may be used in the future to help control a beetle that is



damaging pine trees. The new wasp, *Callimomoides monochaphagae* Yang, is described in an article in Annals of the Entomological Society of America.

Callimomoides monochaphagae is a solitary wasp that parasitizes eggs of the pine sawyer (*Monochamus alternatus* Hope), a longhorned beetle pest that causes damage to pine trees in China and other eastern Asian countries. The pine sawyer beetles aid the spread of tiny nematodes that cause pine wilt disease, which

has killed trees in a large area of China, causing economic and environmental damage.

“*C. monochaphagae* has great potential as a biocontrol agent for suppression of the longhorned beetle,” the authors write.

If they are able to control the beetle, then the



spread of the nematodes and the disease can be controlled as well. Like many parasitoid wasps, *C. monochaphagae* is very small. Females are only 5.7-7.6 mm in length, and the males are even smaller at 2.8-3.2 mm.

The wasp gets its scientific name from the fact that it parasitizes this particular beetle. “*Monochamus*,” the genus of the pine sawyer beetle, plus the latin word “phaga,” which means to eat, are put together to make the species name “*monochaphagae*.”

However, the wasp does not actually eat the beetle. Instead, females locate eggs from pine sawyer beetles and then lay eggs of their own inside of them. When a wasp larva hatches, it eats the contents of the beetle egg and then chews a hole in the egg and exits it.

Designer Insects are the Latest Weapon in Pest Control

September 18 2013 by Andy Coghlan, New Scientist

GM insects are a go. Genetically modified versions of a fly that decimates olive trees could soon be released in Spain in an attempt to control the pests. And in Brazil, GM mosquitoes are already at large as part of the biggest project yet involving engineered insects. The aim is to stamp out their natural counterparts, which carry dengue fever.

The techniques are opposed by anti-GM groups, but advocates say they offer a real alternative to spraying insecticides, and have the potential to provide a better and more precise tool to combat agricultural pests and insects that spread human diseases.

The olive fruit fly is a major pest to olive growers in the Mediterranean and California. Females lay their eggs in the fruit and the larvae destroy 15 to 30 percent of the crop each year as they munch their way to freedom. To tackle this, Oxitec, a biotech company based in Oxford, UK, has submitted an application to the National Biosafety Commission in Spain to release a small contingent of male GM olive flies, in what would be the first field trial of its kind in Europe.

The males are equipped with genes that cause any female offspring they sire to die while they are still in the larval stage, before they have had a chance to reproduce. The idea is that the male offspring inherit the lethal genes and continue to propagate them through the population, until the number of females dwindles to zero and the species crashes.

In lab experiments, the designer flies successfully competed with non-GM males for mates and caused the population to nosedive within 10 weeks (BMC Biology, doi.org/nvd). To determine what happens in the wild, the trial would involve releasing the engineered flies under six olive trees in the Tarragona province, draped in mesh netting to prevent escape.

As the Spanish authorities mull the application, Oxitec is continuing with an even bigger trial to combat dengue fever in Jacobina, a city of 50,000 people in the Brazilian state of Bahia. The aim: to run local mosquitoes that transmit dengue out of town.

The principle is almost the same as with the olive flies – send out an army of male mosquitoes equipped with genes that prevent any offspring, male or female, from reaching breeding age. "Our males are mating at the frequency we'd hoped," says Luke Alphey, co-founder of Oxitec.

In a preparatory field study elsewhere, the team showed its mosquitoes reduced the native

population by between 80 and 96 percent within six months. "The new program is less about 'does this work?', and more about the first operational roll-out of this technology," says Alphey.

The main benefit of the GM insect method, says Tony Nolan from Imperial College, London, is that it is "strictly species-specific" as the insects only seek out mates of the same species. Controlling the pests with insecticides wouldn't be so discriminating.

However, opponents, such as GeneWatch UK, say there are unanswered questions about the insects, whether they will spread unpredictably, for example. They also point out that sites cleared of insects would eventually be reinvaded. Alphey counters that similar work using males sterilized with radiation has shown that reinvasions can be controlled for decades by simply releasing more insects.

Predators Delay Pest Resistance to Bt Crops

March 4, 2014 Cornell University

Crops genetically modified with the bacterium Bt (*Bacillus thuringiensis*) produce proteins that kill pest insects. Steady exposure has prompted

concern that pests will develop resistance to these proteins, making Bt plants ineffective.

Cornell research shows that the combination of natural enemies, such as ladybeetles, with Bt crops delays a pest's ability to evolve resistance to these insecticidal proteins.

"This is the first demonstrated example of a predator being able to delay the evolution of resistance in an insect pest to a Bt crop," said Anthony Shelton, a professor of entomology at Cornell University's New York State Agricultural Experiment Station in Geneva, N.Y., and a co-author of the paper. Xiaoxia Liu, a visiting scientist from China Agricultural University who worked in the Shelton lab, is the lead author on the paper published in the journal PLoS One.

Bt is a soil bacterium that produces proteins that are toxic to some species of caterpillars and beetles when they are ingested, but have been proven safe to humans and many natural enemies, including predaceous ladybirds. Bt genes have been engineered into a variety of crops to control insect pests.

Since farmers began planting Bt crops in 1996 with 70 million hectares planted in the United States in 2012, there have been only three clear-cut cases in agriculture of resistance in caterpillars, and one in a beetle. "Resistance to Bt crops is surprisingly uncommon," said Shelton.

To delay or prevent insect pests from evolving resistance to Bt crops, the U.S. Environmental Protection Agency promotes the use of multiple Bt genes in plants and the practice of growing refuges of non-Bt plants that serve as a reservoir for insects with Bt susceptible genes.

“Our paper argues there is another factor involved: the conservation of natural enemies of the pest species,” said Shelton. These predators can reduce the number of potentially resistant individuals in a pest population and delay evolution of resistance to Bt.

In the study, the researchers set up large cages in a greenhouse. Each cage contained Bt broccoli and refuges of non-Bt broccoli. They studied populations of diamondback moth (*Plutella xylostella*) larvae, a pest of broccoli, and their natural enemies, ladybird beetles (*Coleomegilla maculata*), for six generations.

Cages contained different combinations of treatments with and without predators, and with and without sprayed insecticides on the non-Bt refuge plants. Farmers commonly spray insecticides on refuge plants to prevent loss by pests, but such sprays can kill predators and prey indiscriminately.

The results showed that diamondback moth populations were reduced in the treatment containing ladybird beetles and unsprayed non-Bt refuge plants. Also, resistance to Bt plants evolved significantly slower in this treatment. In

contrast, Bt plants with no refuge were completely defoliated in treatments without ladybirds after only four to five generations, showing rapid development of resistance in the pests. In the treatment with sprayed non-Bt refuge plants and predators, diamondback moth populations were reduced, but the larvae more quickly evolved resistance to the Bt plants.

“These results demonstrate the effectiveness of Bt plants in controlling the pest population, the lack of effect of Bt on the predators and the role predators play in delaying resistance to Bt plants in the pest population,” said Shelton.

Story Source:

The above story is based on materials provided by Cornell University. The original article was written by Krishna Ramanujan. *Note: Materials may be edited for content and length.*

Journal Reference:

1. Xiaoxia Liu, Mao Chen, Hilda L. Collins, David W. Onstad, Richard T. Roush, Qingwen Zhang, Elizabeth D. Earle, Anthony M. Shelton. Natural Enemies Delay Insect Resistance to Bt Crops. *PLoS ONE*, 2014; 9 (3): e90366 DOI:10.1371/journal.pone.0090366

Crazy Ants Dominate Fire Ants by Neutralizing their Venom

Nathan Jones and Lawrence Gilbert, the University of Texas at Austin

AUSTIN, Texas — Invasive "crazy ants" are rapidly displacing fire ants in areas across the

southeastern U.S. by secreting a compound that neutralizes fire ant venom, according to a University of Texas at Austin study published in the journal Science Express. It's the first known example of an insect with the ability to detoxify another insect's venom.

The crazy ant invasion is the latest in a series of ant invasions from the southern hemisphere and, like its predecessors, will likely have dramatic effects on the region's ecosystems.

Known for their painful stings on humans and other animals, fire ants dominate most ant species by dabbing them with powerful, usually fatal venom. A topical insecticide, the venom is two to three times as toxic as DDT on a per weight basis.

When a crazy ant is smeared with the venom, however, it begins an elaborate detoxification procedure, described for the first time in this study. The exposed crazy ant secretes formic acid from a specialized gland at the tip of its abdomen, transfers it to its mouth and then smears it on its body. (Watch a video of the process: <http://youtu.be/CaAq25JQM4k>.)

In lab experiments, exposed crazy ants that were allowed to detoxify themselves had a 98 percent survival rate. This chemical counter-weapon makes crazy ants nearly invincible in skirmishes with fire ants over food resources and nesting sites.

"As this plays out, unless something new and different happens, crazy ants are going to displace fire ants from much of the southeastern U.S. and become the new ecologically dominant invasive ant species," said Ed LeBrun, a research associate with the Texas invasive species research program at the Brackenridge Field Laboratory in UT Austin's College of Natural Sciences.

Last year, the researchers reported that where crazy ants take hold, the numbers and types of arthropods — insects, spiders, centipedes and crustaceans — decrease, which is likely to have ripple effects on ecosystems by reducing food sources for birds, reptiles and other animals. They also nest in people's homes and damage electrical equipment.

LeBrun described watching a battle for food between red fire ants and crazy ants along the boundary between their two populations at a Texas field site. The fire ants found a dead cricket first and were guarding it in large numbers. Usually when fire ants amass around a food resource, other ants stay clear for fear of their deadly venom.



Crazy ants (on the right) coat themselves with formic acid to neutralize the venom of the fire ant (at left).

"The crazy ants charged into the fire ants, spraying venom," said LeBrun. "When the crazy ants were dabbed with fire ant venom, they would go off and do this odd behavior where they would curl up their gaster [an ant's modified abdomen] and touch their mouths."

It was then that LeBrun first suspected the ants were somehow detoxifying the fire ant venom. Experiments back at the Brackenridge Field Laboratory in Austin helped him and his colleagues identify the detoxification agent and measure its effectiveness.

To test the effectiveness of the formic acid, researchers sealed the glands of crazy ants with nail polish and put them in vials with red fire ants. Without the ability to apply the detoxifying compound to themselves, about half of the crazy ants dabbed with fire ant venom died. Among a control group of crazy ants with unsealed glands, on the other hand, 98 percent survived.

Crazy ants and red fire ants are both native to northern Argentina and southern Brazil, where their ranges have overlapped for a very long time. The researchers suggest this newly discovered detoxification behavior is the result of an ancient evolutionary arms race.

It's still not clear how formic acid renders imported fire ant venom nontoxic. One possibility is that it prevents the venom from penetrating the outer layers of a crazy ant's exoskeleton.

Apart from human intervention, said LeBrun, the only thing stopping the relentless march of the crazy ants will be natural factors, such as arid soils or severe freezes that will be too harsh for them to survive. Like the fire ants before them, their range will ultimately be determined by geology and climate.

There is one bright spot for humans. Unlike fire ants, colonies of crazy ants spread very slowly — about 600 feet per year. The only way they can spread long distances is when transported by people in potted plants and recreational vehicles. LeBrun suggested that people not buy plants if they see ants nesting in the pots and that if they live in areas already invaded by crazy ants, they check for stowaways when they move homes or travel long distance.

"If you have an RV, inspect the campgrounds you visit before parking for the night," said LeBrun. "If you live in infested areas, don't store

food in your vehicles and consider treating your camper with insecticides several days before a trip. Consult with a pest control professional as to the best products to use. Not storing food in any vehicle parked in an infested area is also a good idea."

Flight of the Living Dead: 'Zombie' Bees Surface in the Northeast

Thu, Jan 30, 2014 by BEN GITTLESON, ABC News

Mutant "zombie bees" that act like the ghoulish creatures of horror films have surfaced in the Northeast after first appearing on the West Coast.

An amateur beekeeper in Burlington, Vt., last summer found honeybees infested with parasites that cause the insects to act erratically and eventually kill them. It was the first spotting of zombie bees east of South Dakota, according to John Hafernik, a professor of biology at San Francisco State University whose team in October verified the infestation.

"They fly around in a disoriented way, get attracted to light, and then fall down and wander around in a way that's sort of reminiscent of zombies in the movies," Hafernik said. "Sometimes we've taken to calling [it], when

they leave their hives, 'the flight of the living dead.'"

The professor accidentally discovered the zombie bees in California in 2008, and since then cases have been reported in Oregon, Washington state, California and South Dakota, he said.

The effect starts with a fly called *Apocephalus borealis*, which latches onto European honeybees — common across the United States — and lays eggs in the bees that eventually hatch and wreak havoc on their hosts, Hafernik said.



"It's sort of a combination of zombies and aliens mixed together," he said.

But there's not necessarily any threat of a zombie (bee) invasion anytime soon, according to Hafernik.

The Vermont iteration of the bees first came to light when Anthony Cantrell, a hardware-store employee who took up beekeeping as a hobby less than a year ago, noticed some dead bees

outside his home. Later, he came across ZomBeeWatch.org, a website run by Hafernik and his colleagues, and realized some of his bees might have become infested.

Honeybees sometimes become infested by other parasites and diseases. "I just thought, great, one more thing that the poor honeybee has to deal with," Cantrell, who has two hives, told ABC News.

Steve Parise, an agriculture production specialist with the Vermont Agency of Agriculture, Food and Markets, on Tuesday brought up the threat posed by zombie bees, at a meeting of the Vermont Beekeepers Association, according to Cantrell, who is a member.

Farmers rely on bees to pollinate agriculture fields and produce honey, but there hasn't been any sign of a widespread infestation, even though it remains unclear just how many bees across the continent have been infested, he said.

While researchers at San Francisco State University have confirmed reports of the parasitic flies infesting bees up and down the West Coast — one-third of hives in the San Francisco Bay Area may at certain points in time be infested — no confirmed cases have popped up in the Northeast since October, according to Hafernik.

Once the flies infest bees with their eggs, the bees start exhibiting zombielike behavior; then,

once the eggs hatch, they generally drop dead after about five minutes, he said.

The culprit fly was originally discovered in the 1920s, in Maine, and has been found across the United States, where it had been known to parasitize bumblebees and yellow jacket hornets — but not honeybees, he said.

In Vermont, the state's Agency of Agriculture may trap bees to investigate the zombie bee threat, according to The Associated Press.

Cantrell's waiting out the winter to see if the parasite survives the winter.

Forest Pest Management Cooperative's

P.E.S.T. Newsletter

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