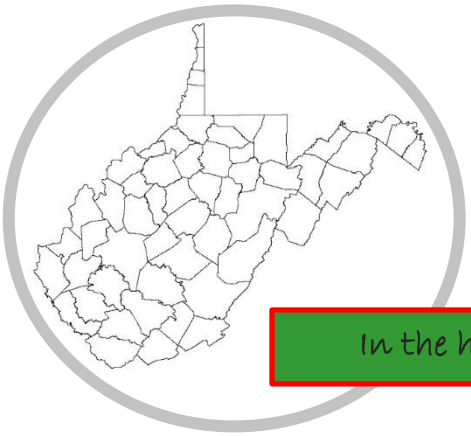

The West Virginia Chapter of The American Chestnut Foundation

NEWSLETTER

In the heart of American chestnut's natural range

October 2020

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Invasive Species

The goal of TACF is to restore American chestnut into the forests of Eastern North America. There are many reasons to restore this magnificent tree, but one reason is that American chestnut in a native plant. You don't have to look too far in West Virginia to see many invasive plants—kudzu, Autumn olive, Tree-of-Heaven, multiflora rose, Oriental bittersweet, and Japanese honeysuckle to name a few. An article in the April 2020 edition of ***Smithsonian*** magazine was written about **Dr. Douglas Tallamy**, an ecologist who urges Americans to plant native species in their yards and forests. Tallamy wants America replanted with North American flora, supporting a healthy array of native butterflies, moths and arthropods, providing food for a robust population of songbirds, small mammals and reptiles. Tallamy, an entomologist at the University of Delaware, was struck by the absence of caterpillars on his property in southeastern PA. Chickadees can consume 6,000 to 9,000 caterpillars before they leave the nest, all within a 150-foot radius of their nest. Currently, the birds would have a hard time finding food in his part of PA, because the plants on his property were mostly invasive species, either planted or brought to America accidentally. The invasives escaped and outcompeted their native counterparts. By and large, most plants can tolerate a wide range of environmental conditions. But insects tend to be specialists, feeding on and pollinating a narrow spectrum of plant life, sometimes just a single species. Tallamy states that "90% of the insects that eat plants can develop and reproduce only on the plant with which they share an evolutionary history". Plants have developed various chemical and morphological defenses--toxins, sticky sap, waxy cuticles, thick bark--and insects have found a way to get around them. However, insects don't work well against species they have never encountered. This can occur even on closely related species like Norway and sugar maples. Tallamy found that introduced plants, on average, provide 68% less food for insects than native species. Thus, a native plant may harbor hundreds of insects, supporting birds and small mammals that invasive species do not support.

Tallamy stated that one-third of all vegetation in southeastern PA is from Asia. Invasive species are called that for a reason and repelling them is hard and never-ending work.

Not all native plants are created equal, at least from the point of an insect. Across a wide range of North American biomes, about 14% of plants make up 90% of the insect food. These are the keystone species that keep the food web healthy and the most important are four genera of native trees: oaks, poplars, willows and cherries. But also hickory, chestnut, elms and birches.

It is possible that chestnut would have been one of the keystone species prior to the arrival of another invasive, the chestnut blight fungus.

Keystone species are important as food sources for insects. When insects disappear, humans may not take much notice, but the recent population declines of two species have received a great deal of attention: the monarch butterfly, because it's an iconic, easily recognizable and beautiful creature; and the honeybee, because it's need to pollinate crops. But those two episodes are symptomatic of a larger disruption in the ecosystem. Tallamy estimates that the worldwide population of arthropods, chiefly

insects, has declined 45% from preindustrial times. Without insects, it would be that case that lizards, frogs and toad, birds and mammals, from rodents up to bears, would lose all or a large part of their diets. The little things in the world are disappearing.

The world-wide food web is very interwoven. Wood lice feed on decaying stumps and the lice are food for spiders, frogs and bird. Tallamy quoted noted conservationist, E.O. Wilson, from his 1987 talk, *"The Importance of Conservation of Invertebrates"*. Wilson said, "The truth is that we need invertebrates, but they don't need us. If human beings were to disappear tomorrow, the world would go on with little change…But if invertebrates were to disappear, I doubt that the human species could last more than a few months. Most the fishes, amphibians, birds and mammals would crash to extinction about the same time. Next would go the bulk of the flowering plants and with them the physical structure of the majority of forests and other terrestrial habitats of the world. The Earth would rot".

The bottom line, plant native plants, including American chestnut.

Read the article in its entirety: Wild Man by Jerry Adler, Smithsonian, April 2020, pages 80-94.

Recap of WV Chapter Meeting

A virtual chapter meeting was held Saturday, October 10 with 20 WV chapter members participating. The meeting minutes follow.

- The meeting minutes from the 4 April 2020 meeting were approved.
- The treasurer's report, presented by Sam Muncy, was approved. The general account balance is $32,952.94 and the Waddell account balance is $38,573.05 for a total of $71,470.22.
- WV-TACF summer intern, **Logan Hosaflook**, a senior forestry student at Glenville State College, submitted a report on his summer activities.
- **Tom Saielli**, TACF's mid-Atlantic regional science coordinator gave a presentation.
  - The transgenic tree (OxO) is one of many tools that some state chapters (with federal permission) are incorporating into their plantings. If the OxO tree is deregulated, those trees can then be crossed with the backcross material from TACF as well as native American chestnut trees. Subsequent crosses will breed out all the but the OxO gene, providing states will locally adapted, blight tolerant chestnut trees. Thus, WV needs more germplasm conservation orchards (GCOs). *See article on Page 7*.
  - USDA-APHIS has provided some states permits to use OxO pollen to pollinate American chestnut trees.
  - The public comment period for the OxO tree closes Oct. 19, 2020. To date, positive responses have been about 70%. If USDA-APHIS deregulates the OxO tree, then the EPA and FDA will likely follow.
  - Some OxO tree have been placed under high light and they produce copious amounts of pollen at a young age. Tom was able to use the OxO pollen and pollinate about 15 American chestnut trees. Thus, the first backcross trees with OxO pollen are in the greenhouse and they can be outplanted if deregulated.
  - Grafting scion is fickle. Some grafts take and some do not. Several WV-TACF members collected scion wood in 2020 that was shipped to Dr. Hill Craddock at the University of TN-Chattanooga. With Covid-19, Hill had little help, as students were not permitted on campus, so Hill was unable to graft as many scions as he hoped. Tom asked Hill to prioritize the WV scions. A grafting workshop led by Hill Craddock was scheduled for 2020, but it had to be canceled due to the pandemic.
  - Root stooling is another way to produce American chestnuts. Tom detailed the steps necessary to get a young root sprout to develop roots that can then be transplanted.
    - Get permission from land-owner if necessary.
    - Collect detailed data: location, county, TreeSnap…etc.
    - Use young stump sprouts and not epicormic shorts. Young sprouts have a lot of

undifferentiated cells that ultimately can become shoots or roots.

- Supplies include: 18-gauge wire; pliers; rooting hormone, either powder or liquid; paint brush to apply hormone; flagging; hand towel and bucket, and water.
- Technique is to pull the wire tightly around the sprout. This will force root formation. Make a coil of wire, paint on the hormone about 1-1.5" and then pack soil around the base of the sprout.
- The proof of the success of this technique will be a year from now when the roots can be dug up and replanted successfully.

- Tom reported that TACF is working to answer the question, "is there a tradeoff between blight resistance and the percentage of American germplasm—how do we balance 'resistant enough' with 'American enough'"? We need to know what percentage of Chinese DNA can be allowed in advanced TACF backcross hybrids and still replace the ecological niche once occupied by American chestnut. They are comparing ecological fitness among various hybrid genotypes. SUNY has conducted a number of ecological studies in NY (ie herbivory, honeybee impact, decomposition of leaf litter, etc). Currently, such studies being conducted in KY and PA.

- **Robert Sypolt** and **Darrell Dean** reported on their work around the state. Darrell wrote an article for TACF's *Chestnut* magazine about large surviving American chestnuts in Hardy and Pendleton counties. **Jim Bowen**, WV forester for Hardy and Grant counties, has helped Robert and Darrell locate a number of surviving chestnut trees and Jim is collecting nuts from these trees for GCOs in the state. Robert collected about 150 nuts from the Waddell orchard at Preston County High School. Robert normally has a field tour of the Waddell orchard with students from Preston County schools, but due to the pandemic, he made a video that can be presented to students. Robert also developed a chestnut trail for the cross-country teams. Darrell highlighted the Sandra Wales orchard in Preston County. He and Robert began planting trees from the Clements tree nursery at this site in 2013 and additional trees have been planted in subsequent years. To date, the survival rate in 65%.

- **Dr. Melissa Thomas Van Gundy** asked that members use the TreeSnap app when locating native American chestnut trees. She is working on two plantings in the Monongahela National Forest and she hopes to have the data published in 2021. The stands are in Cowen and St. George. She provided a link to a paper on chestnut being a fire-adapted species: https://esajournals.onlinelibrary.wiley.com/doi/10.1002/ecs2.3267
  She also provided a link to a paper on the comparison of the past forest of the Monongahela (and private land within) and the current forest. https://www.nrs.fs.fed.us/pubs/61224.
- **Bolgiano Property** in Randolph County was discussed. **Betsy Gamber**, TACF VP, sent an email stating the Foundation was considering selling the property. On October 2, **Rick Sypolt** and summer intern **Logan Hosaflook**, painted the boundary of the property with purple paint. Rick was in favor of leasing the property to a hunting group as the hunters could be asked to oversee a chestnut planting as part of an agreement. Boundary painting will lessen liability issues since the boundary is now well defined. Rick reported that he did not see as much 4-wheeler activity on the property as he did in 2019. **Jimmy Jenkins** surveyed the property and valued the timber at about $40,000. Jimmy also suggested we keep the property as there is marketable timber. Jimmy did suggest that with a depressed timber market, any timber sale should be delayed until timber prices increase. It was unknown if taxes are paid on the property. It was suggested that the chapter could pay the taxes if

necessary. Mark Double was asked to write to Betsy Gamber and express the chapter's wish to keep the property.

- **The Boy Scout Jamboree** at the BSR has been canceled for 2021 due to Covid-19. A work date at BSR is scheduled for October 24. Contact Sam Muncy if you can assist (sam.muncy@msesinc.com).

- **Clements Tree nursery** in Mason County, may be closing. The entire nursery is managed by 1.5 employees; however, the part-time position will not be filled as of Jan 2021. Much of the seedling lifting and packaging is assisted by female inmates at the Lakin Correctional Center and Jail, a facility adjacent to the nursery. Due to Covid-19, the inmates have been forbidden to work. The nursery normally sells 400-500K seedling each year, but they were not provided funds to purchase seeds this fall. Thus, the nursery has only 40,000 seedlings available for sale in 2021. The nursery annually sells 5,000-10,000 American chestnut seedlings and closure of the nursery would greatly impact the chapter as many members have trees that they purchased from the nursery. **Dr. Joe Golden**, spoke to **Ed Gaunch**, WV Secretary of Commerce and found out that nursery sales have decreased due to the fact that one of the largest customers, an out-of-state contractor, no longer purchases seedlings. Joe suggested an advisory committee might be able to assist the nursery. The committee could be comprised of the WV Dept. Ag., Division of Forestry, Master Naturalists, Master Gardeners and someone from the business community. Letters of support would be welcomed from WV-TACF members. Email **Tom Cover** (C.Tom.Cover@wv.gov) Director of the WV Division of Forestry and **Ed Gaunch** (Ed.Gaunch@wv.gov) to encourage them to keep the nursery operational.

- Board members and officers were approved for their respective terms (2 years for President, VP, Secretary and Treasurer) and 3 years for Board members.

- GCO's for 2021 were discussed. It appears that 2020 is our best year as a chapter collecting chestnuts from native American trees. Possible GCO sites include:
    - Riggleman farm in Franklin
    - Sutton Dam
    - Terra Alta property owned by Joe Nassif
    - Twin Falls State Park
    - Oak Hill school complex

Biological Control

Several of the recent WV chapter newsletters have featured the transgenic tree developed by the State University of New York. The transgenic tree falls used "Biotechnology", one of TACF's three approaches (***3BUR*: Breeding, Biotechnology and Biocontrol United for Restoration**) to create a disease tolerant and genetically diverse population of American chestnut that will be adaptable to broad and changing climate.

The focus on this article will be on **Biocontrol**, short for biological control (the control of a pest by the introduction of a natural enemy or predator).

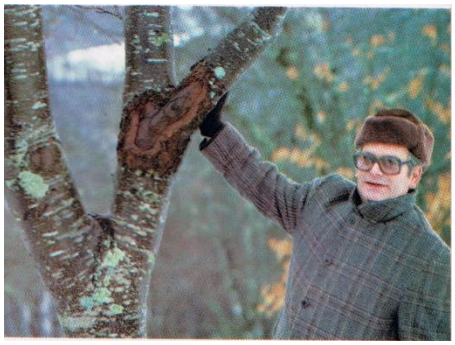
The story of biocontrol begins long before TACF was founded (in 1983). The chestnut blight fungus attacked and killed not only American chestnut (*Castanea dentata*), but also European chestnut (*Castanea sativa*). While chestnut blight was first noticed in the U.S. in 1904, the fungus was first found in Europe in 1938 in Genoa, Italy. The blight quickly spread to Spain, France, Greece, Turkey and Switzerland. Unlike American chestnut that grew straight and tall in the wild and was utilized mostly for nut production and timber, European chestnuts were grown mostly in orchard settings for nuts. European chestnuts were known as the **Bread Tree**, and its nuts were used as a food source since the Middle Ages. Chestnuts were roasted in huge numbers laid upon thin hazel branches over hot coals and they were then ground down into flour in order to make a kind of polenta. The flour could also be used to make bread, in the absence of grain flour.

Antonio Biraghi, a Swiss plant pathologist, was one of the first to notice a change in the disease on European chestnuts. In the 1950s, Biraghi was walking through abandoned European chestnut

orchards in Switzerland, when he noticed that some of the trees were alive with rough bark and callousing canker.

Superficial cankers on European chestnut observed by Biraghi.

Upon further examination, Biraghi hypothesized that the tree had mutated. It turned out that Birgahi's hypothesis was incorrect. Biraghi summoned the help of a mycology graduate student in France, **Jean Grente**. Together, Biraghi and Grente examined the non-killing cankers on Swiss European chestnuts, and Grente took a number of bark



Jean Grente, French mycologist, examines a canker on European chestnut.

samples from the cankers. When Grente returned to his laboratory in France, he expected to culture the typical orange-pigmented chestnut blight fungus. Much to his surprise, Grente isolated a fungus with much less orange pigment—the culture was nearly white.

Two cultures of the chestnut blight fungus. The orange-pigment isolate (left) is pathogenic and kills chestnut, while the white isolate (right) contains a virus and is much less pathogenic.

Grente began year-long studies with the orange- and white-pigmented isolates, conducting both laboratory and field studies. Grente inoculated both isolates into European chestnut trees and found that while the orange-pigmented fungus girdled and killed a stem, the white-pigmented fungus grew a little in the tree and then stopped. Grente termed the white-pigmented fungus, hypovirulent (hypo means "less than"), since the white-fungus produced cankers that were less virulent than the orange-pigmented isolate.

Chestnut stems inoculated with the white-pigmented isolate (top) and an orange-pigmented isolate (bottom). The white-pigmented isolate produces a much smaller canker.

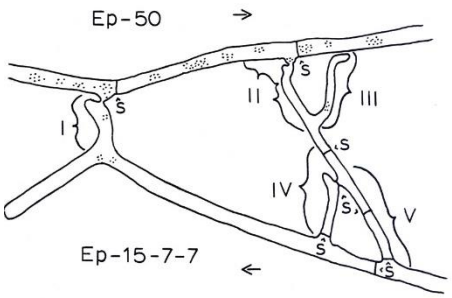
What is hypovirulence and how does it work?

The slow growth of the hypovirulent isolates is the result of infection of the fungus by a virus. It seems like viruses are a current focus concerning human health with Covid-19 and annual influenza. I've used **Roger Bannister** as a human example to explain the effect of a virus.

Bannister was the first human to run a sub-4 minute mile. Thus, a healthy

Roger Bannister became the first man ever to break the 4-minute mile May 6, 1954 in Oxford, England.

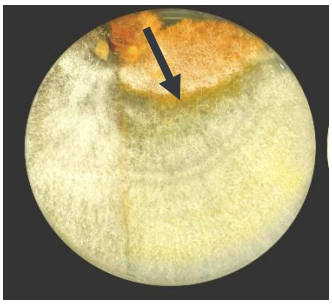
Bannister is analogous to the orange-pigmented fungus, as the fungus is able to grow, girdle and kill American chestnut trees. What if Bannister was infected with a stomach flu virus that caused a high fever, nausea, vomiting and diarrhea? He might be able to run a mile, but in his weakened condition, it might take him 30 minutes or longer. The stomach virus doesn't' kill Bannister, but it weakens him. The same holds true for the virus that infects the chestnut blight fungus, in that the fungus is weakened but not killed. The virus disrupts the metabolism of the fungus such that it produces less pigment. In addition, the white-pigmented fungus sporulates less and is less able to grow in chestnut bark.



The dots in the upper strand indicate virus infection. When the fungal strand below fuses with the virus-infected strand, the cytoplasm of the two mix and virus is transported to the formerly uninfected stand.

In the above cartoon, there are strands of two different isolates of

the chestnut blight fungus. The upper strand (white-pigmented fungus) contains virus (indicated by the dots). The lower stand is attracted to the upper strand and the two fuse. As a result, there is mixing of the cytoplasm (the liquid material in a living cell, excluding the nucleus) and the virus moves from the white-pigmented strand to the orange-pigmented strand. The orange-pigmented strand becomes infected and turns white as seen in the photo below (black arrow).



Two isolates of the chestnut blight fungus (white- and orange-pigmented) are paired side-by-side on an agar Petri plate. After 3-4 days of growth, the orange-pigmented isolate fuses with the white-pigmented isolate and the virus is transmitted through the mixing of cytoplasm. As a result, all subsequent growth of the orange-pigmented isolate is white, meaning it is virus-infected.

How are hypovirulent isolates used to treat cankers?

Hypovirulent isolates are grown in the laboratory and mixed with agar and water to make a thick slurry, the consistency of applesauce. This process is done in a sterile environment so not to introduce contamination to the slurry.

Many plates of hypovirulent isolates are used to make a slurry.

Holes around the perimeter of a canker are filled with hypovirulent inoculum.

Wounds are made around the perimeter of a chestnut blight canker and the slurry is added to the wounds. If successful, the former virulent fungus acquires the virus particles, its growth is slowed so that the tree can use its natural defense mechanisms and produce callus tissue to wall off the fungus.

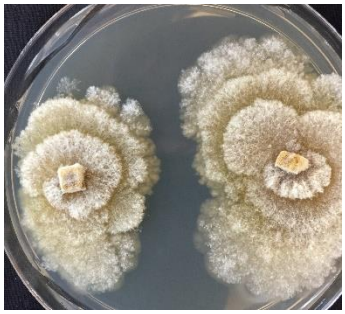
Callus tissue is formed by a tree after a canker is treated successfully.

The problem with traditional hypovirulent isolates is they do not interact with all 64 known types of the chestnut blight fungus.

Testing the compatibility of isolates of the chestnut blight fungus. There are five pairings on the plate above. Only the pairing (on the lower right) is compatible. The other four pairings indicate that the two isolates are incompatible.

Most traditional hypovirulent isolates (those found in nature) will pass along the virus to only a few of the 64 compatibility types. In chestnut forests in WV, there can be as many as 25-30 compatibility types in a small area, making the fungus highly diverse. Thus, finding the exact match of the fungus in a natural canker with a hypovirulent isolate is less than ideal, so biological control in the forest is difficult.

To combat the complexity of the 64 types, **Dr. Donald Nuss**, University of Maryland (retired), developed viral strains that interact with all 64 types.



Super donor isolates in culture.

Nuss refers to these isolates as **Super Donors**. This method can keep blight-susceptible American chestnut trees in germplasm conservation orchards alive and healthy so that they can be used in our transgenic and traditional breeding programs.

Don Nuss applied to USDA-APHIS to have the super donor isolates deregulated—somewhat similar to the application by the State University of New York to have the transgenic trees deregulated. Nuss was recently informed that the super donor isolates are still considered plant pests, so each application of the strains (orchards or forests) will have to have a USDA-APHIS permit.

Landscape Cloth

An article from the 26 Sep 2020 edition of the ***Chicago Tribune***, written by **Beth Botts** of the Morton Arboretum, detailed potential problems with landscape cloth. **Sharon Yiesla**, plant knowledge specialist at the Morton Arboretum, stated that the fabric may lead to problems. For example, it is designed to allow passage of both water and air, which are critical for the roots of trees and other plants. Often there are small perforations for this purpose. But in practice, the openings may clog up with soil and organic matter from the decay of mulch. Air can't get through and neither can rain. Yiesla indicated that with landscape cloth, you could unknowingly create a desert for your plant. They may be living in drought conditions, even if you water, because the water cannot reach the soil and the roots. Yielsa also stated that even with mulch on top of landscape cloth, nutrients from decaying mulch are blocked from improving the soil. The suggestion was to simply use an even layer of mulch over the soil around trees, 3"-4" deep at least 3' out from the trunk.

Germplasm Conservation Orchard Plans for 2021

The WV chapter has big plans for next year, as we hope to install 4-5 germplasm conservation orchards (GCOs). Each orchard will ultimately have 100 American chestnut trees (10 trees each from 10 different WV sources). We will not have 10 sources of nuts next year, but we hope to start the orchards with 3-4 sources (or 30-40 seedlings). West Virginia is the only one out of 16 state chapters that does not have dedicated CGOs. Some states have 15-20 GCOs that are more than a decade old**. Why do we need orchards of pure American chestnut? Won't they die from chestnut blight?** The short answer is that American chestnut trees are highly susceptible to the chestnut blight fungus. While all the trees will eventually contract the disease, it has been shown and about 50% of the trees will resprout and the new sprouts will grow sufficiently to produce flowers. It is the flowers that the chapter will need to hybridize with TACF's advanced backcross trees or the transgenic chestnuts produced by the State University of New York, if the tree is deregulated by USDA-APHIS. Using pollen from native WV trees will be the first step in the restoration process, as trees adapted to WV can be planted across the state. This will be a big step for the WV chapter. The reason for giving you advance notice is that installing GCOs will require many hands. Each plot will have to be laid out, holes dug, trees planted, cages cut and installed, mulch applied and the seedlings watered. Even though we might only be planting 40 trees per site, the more hands we have the easier the process. Times and dates of the GCO installations will be announced in the spring of 2021. It is hoped that many WV chapter members will be able to assist with this vital project.

Spotted Lanternfly

Spotted lanternfly (*Lycorma delicatula*) is an exotic and invasive insect from China that has the potential to spread rapidly throughout West Virginia. This invasive hemipteran is known to severely damage both the forestry and agricultural industries in states with high infestations. Both the grape and fruit crop industry and timber production of our native forest trees are greatly impacted by the feeding habits of large populations of spotted lanternfly. Spotted lanternfly is a sap feeding insect, which stabs its proboscis mouth part into the tree or vine it is infesting and sucks out necessary water and sugars the tree needs to grow fruit and maintain a healthy canopy. When hundreds of spotted lanternflies all feed on one tree or vine it can greatly reduce the yield of the tree and eventually the tree or vine will be so weakened that it can die if the spotted lanternfly population is not controlled. Additionally, the excretion of honeydew by the spotted lanternfly leads to large thick black mats of sooty mold on foliage under the infestation. The sooty mold can be so thick that understory branches of the host plant itself and additional understory shrubs can die from the lack of ability to perform photosynthesis. The monetary and ecological impacts of this insect are very high in the state of West Virginia.

Spotted lanternfly overwinters as eggs, in a gray putty-like egg mass. Egg masses are laid on smooth surfaces of host plants and also man-made items like vehicles, trailers, outdoor equipment and patio furniture. This habit of laying eggs on movable surfaces allows the insect to spread rapidly. Eggs

hatch in spring. Nymphs (immature insects) are wingless and can be found in spring through early summer. Young nymphs are small and black with white spots. When the nymph matures and molts it becomes bright red with white spots. After two additional molts the red nymph will molt into a large (~1 inch) winged adult in late July. The adults congregate on trees, usually on its preferred host tree-of-heaven (*Ailanthus altissima*), to feed and mate from late July to October. There is one generation of this insect per year. Female spotted lanternfly on average lay 35 eggs at a time and will lay eggs three separate times as an adult during the months of September-October. Spotted lanternfly is known to feed on over 100 species of trees and has been seen with preferences to birch and maples species when tree-of-heaven populations are low or competently covered in spotted lanternflies, forcing the insects to move to other hosts.

In 2019, a small population of spotted lanternfly was found in Bunker Hill, WV in Berkeley County. In August of 2020, the West Virginia Department of Agriculture and the USDA APHIS discovered a population of around 100 spotted lanternflies near Interstate 81 in Inwood, WV; about 2 miles from the sighting last year. Currently, in October of 2020, West Virginia has two counties infested with spotted lanternfly: Berkeley and Mineral.

"The WVDA is encouraging landowners to inspect their property for egg masses and adult life stages, especially for properties that contain numerous Tree-of-Heaven," said WVDA Spotted Lanternfly Coordinator **Dr. Kristen Wickert**. "The greater the effort to track the movement of this pest, the more effective our treatment can be to combat it. We rely heavily on the public to aid us on this effort."

Egg masses of spotted lanternfly.

Adult spotted lanternfly.

Adult spotted lanternfly with closed wings.

Mass of adults on a tree host.

Current control methods for the spotted lanternfly are physical trapping with the use of sticky bands or circle traps, outreach to the public, expansive visual surveys across the state, mapping of the insect's preferred host tree-of-heaven and treatment with both systemic and contact pesticides depending on the time of the year. WVDA is working with USDA-APHIS to control this pest in West Virginia.

The above article was supplied by Dr. Kristen Wickert, WV Department of Agriculture.

Future Articles

If there are articles that you would like to see in future newsletters, contact the editor, Mark Double @mdouble1@hotmail.com.

WV-TACF Officers

(Elected in October 2020)

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