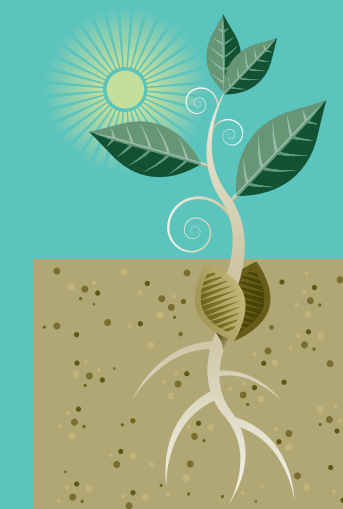


Bulletin news from the experiment station associates



The *Bulletin* is published by the Experiment Station Associates and mailed to our members. To become a member, please complete and mail the form on page 6.

The Experiment Station Associates was formed in 1990 to encourage and support the work of the scientists at The Connecticut Agricultural Experiment Station.

We welcome your comments. Please email the editor, Pamela Weil, at pamelaweil44@gmail.com.

Climate Change and Mosquito-Borne Diseases in the Northeast

by David Yih

At April's annual meeting of the Experiment Station Associates (ESA), Dr. Theodore G. Andreadis, newly appointed Director and former Chief Scientist and Head of The Connecticut Agricultural Experiment Station's Center for Vector Biology & Zoonotic Diseases and Environmental Sciences Department, took up the topic of arthropod-borne viruses (arboviruses), focusing specifically on the effects of climate change on mosquito-borne diseases in the Northeast.

Five years later, an outbreak in Massachusetts killed 25 of the 34 humans who contracted the disease. Similarly, subsequent EEE appearances have tended to alternate with absences of several years, but, as Andreadis and CAES Associate Scientist Philip Armstrong warned in a May 2013 article in the *New England Journal of Medicine*, "Over the past decade we have witnessed a sustained resurgence of EEE virus activity within...the northeastern United States and northward expansion into regions where the virus was historically rare or previously unknown, including northern New England and eastern Canada."

Some people have predicted that global warming will create favorable habitat for mosquitoes and spread tropical diseases into our state. Deadly outbreaks of malaria and yellow fever, spread by sailing ships, once ravaged the eastern seaboard, including many towns in Connecticut and other New England states. But Dr. Andreadis doubts that these will return to Connecticut in the near future. For now, he is more concerned with deadly pathogens that are showing up with increasing regularity here, in particular the eastern equine encephalitis and West Nile viruses. Mosquitoes transmit both diseases.

Eastern Equine Encephalitis (EEE)
EEE is the deadliest mosquito-borne virus in North America, with a 30% mortality rate and long-term damage experienced by 90% of survivors. It is endemic in the eastern U.S. where it was first discovered in 1933.



Aedes japonicus is an invasive mosquito species from Asia.

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("direct photolysis"), but is accelerated by dissolved natural organic matter (DNOM) ("sensitized photolysis") due to energy transfer. Salts inhibit direct and indirect photolysis. Thus, as a parcel of water containing estrogen moves through the salinity gradient from freshwater to seawater, overall photolysis will decline due to the salts effect and DNOM photobleaching, and indirect photolysis will decline relative to direct photolysis. (Environmental Science & Technology 46:7128-7144, 2012).

Plant disease epidemics are often characterized by the latent period (period of time between infection and the initial production of spores) and the infectious period (the period of time over which spores are produced). **Dr. Frank Ferrandino** has shown that this approach can lead to inconsistencies due to the natural variability among infections and that time scales based on the total population of lesions are superior in predicting the resultant speed of an epidemic. (Phytopathology 102 [8]: 728-732).

Craig Musante and Jason C. White have found that nanoparticles such as silver and copper displayed significantly greater phytotoxicity to squash plants grown hydroponically than did the equivalent bulk materials. Conversely, gold and silicon displayed toxicity that did not vary with particle size. In addition, solution conditions such as the presence of humic acid also differentially impacted bulk and nanomaterials with respect to physical and biological activity. These findings are significant because current regulatory guidelines assume nanoparticles and corresponding bulk elements have equivalent toxicity. (Environmental Toxicology, December 2010).

Drs. Wade H. Elmer and Robert E. Marra have described a new species of *Fusarium* that they named *Fusarium palustre* (from Latin palus, referring to the marsh habitat in which this fungus is found). The fungus is moderately pathogenic on marsh grass, *Spartina alterniflora*, and is found associated with plants in areas where Sudden Vegetation Dieback occurs. (Mycologia 103:806-819, 2011).

Neil Schultes and 5 others discovered that genetic and molecular characterization reveals a unique nucleobase cation symporter 1 in *Arabidopsis*. (FEBS Letters 586:1370-1378).

Boxwood Blight Can Infest Pachysandra

The first report of a natural infection of pachysandra (*Pachysandra terminalis*) in the landscape by *Colonectria pseudonaviculata* (syn. *Cylindrocladium pseudonaviculatum*), the boxwood blight fungus, was confirmed by



Photo by Dr. Sharon Douglas

Station plant pathologists in June 2012.

The residential property had installed boxwood-blight infected boxwood plants in May 2012 and the pathogen spread to established pachysandra beds on the property.

Symptoms on the pachysandra included necrotic lesions with distinct margins and diffuse yellow haloes on leaves. Stem lesions can develop, but usually do not result in significant damage.

This follows the first report of pachysandra as a host of boxwood blight by CAES, which involved experimental inoculations conducted in the laboratory and greenhouse. Since the 2112 report, additional natural infections of pachysandra in the landscape have been confirmed in Connecticut.

SCIENTIFIC DISCOVERIES

Sudden Oak Death Found on Rhodie

In July 2011, there was unfortunately a positive find of *Phytophthora ramorum*, the causal agent of Ramorum Blight (or Sudden Oak Death) in a residential property in New Haven County. This was detected through the Station's participation in a "trace-forward" survey initiated by the USDA-APHIS-PPQ on plants sent by mail to private residences throughout the U.S., including Connecticut.

Plants were from a nursery in Oregon that had tested positive for *P. ramorum*. Samples were collected by **Dr. Victoria Smith (Deputy State Entomologist)** and nursery inspectors, and brought to the lab for testing by **Drs. Sharon M. Douglas and Robert E. Marra**. Samples from *Rhododendron* sp. 'Pronum' tested positive for *P. ramorum* using real-time PCR and by culture. Follow-up surveys of host material and soil on the property are in progress.

Jason C. White, Roberto De La Torre Roche and Joseph Hawthorne, along with collaborators at 3 other institutions, have discovered that carbon nanomaterials (fullerenes) added

to the root zone of plants can influence the availability of other organic chemicals. Specially, fullerenes were shown to increase the accumulation of weathered DDE, an estrogenic metabolite of DDT, by tomato, zucchini and soybean. Fullerenes are being considered as additives to pesticides and fertilizer formulations; the discovery that these nanomaterials impact co-contaminant uptake may present concerns over food safety.

Brian Eitzer, along with colleagues at Purdue University, have been studying neonitinoic pesticides in various types of samples taken near cornfields. They found low levels of these pesticides in the soil, dandelions, honey bee collected pollen, and honey bees. Higher levels of these compounds were found in some of the exhaust material produced during corn planting. The results suggest that there are possible unexpected routes of exposure of honey bees to pesticides.

Purple marsh crabs (*Sesarma reticulatum*) are herbivores that are found in high densities in Sudden Vegetation Dieback (SVD) sites. **Dr. Wade H. Elmer** reared purple marsh crabs in captivity and

Continued on next page

demonstrated that they preferred to feed upon drought-stressed, diseased *Spartina* plants more than healthy plants. These findings may suggest that the marsh crabs are attracted to SVD sites after a stress event.

Dr. Neil McHale has been working in collaboration with Dr. Million Tadege (Oklahoma State University) on genes governing the formation of leaf blades, the living solar panels that capture light energy for photosynthesis. The main emphasis is on WOX genes, a highly diverse family of transcription factors with critical roles in the development plant leaves, flowers, roots, embryos and ovules.

Their recent studies show an extensive functional overlap between recently evolved WOX genes and those that existed in primitive tracheophytes over 400 million years ago.

WOX genes were apparently at work guiding the course of plant evolution as soon as simple aquatic plants first ventured out of the oceans and continue to shape the patterns of growth and development in the complex terrestrial plants of today.

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fatal. In the Northeast, only Maine and Connecticut have not had a human case as yet.

West Nile Virus (WNV)

In the spring and summer of 1999, WNV made its first known appearance on the continent in New York. CAES scientists were the first in North America to isolate the virus in the lab. Over the decade and a half since then, 1.6 million people have been infected with WNV in the U.S. alone. Though most do not develop symptoms, among those who do the disease has been fatal in over 1,500 cases.

2012 was a record year for WNV in Connecticut and also saw the highest number of cases ever recorded for New England as a whole.

Normally it takes some time for the virus to build up or “amplify” in an area. As the season advances, more and more birds are infected by mosquito species that specialize in avian hosts, until other mosquito species — “bridge species” that attack both birds and mammals — begin carrying the virus in significant numbers and transmitting it to humans. Conditions in 2012 were

ideal, supporting both earlier emergence and earlier peaking of the virus

Temperature

The current trend toward milder winters, hotter summers, and precipitation extremes in the Northeast has been favorable for many mosquito species. Dr. Arendreis pointed out that although there can be increased mosquito mortality at very high temperatures, warmer temperatures generally favor disease prevalence, in a variety of ways.

Warmer temperatures prolong the mosquito season and extend the geographical distributions of species with high optimum temperature ranges, including tropical species such as *Aedes aegypti* and *A. albopictus*. A longer season means there is more time for the viruses to build up in the avian and mosquito populations as well as more opportunity for human exposure. Milder winters increase the likelihood that infected mosquitoes will survive the winter and jumpstart the disease cycle at a higher initial level the following spring.

Warmer temperatures also have physiological impacts, shortening larval development time and allowing female mosquitoes to feed more frequently, digest blood more rapidly, and produce more offspring.

And increased temperature affects what scientists call the extrinsic incubation period — the time it takes from the ingestion of an infectious blood meal until the insect can transmit the disease causing microorganism to a new host.

Dr. Arendreis noted that while some viral particles are destroyed in the mosquito’s digestive process, others pass intact through the semi-permeable membrane that surrounds the mosquito’s digestive tract and migrate to various areas within the mosquito’s body. Those that show up in the salivary gland will infect the next host the insect bites. This extrinsic incubation period is shorter in warmer temperatures.

Monitoring Insects Before They Arrive by Robert Pollack

Although the brown, marmorated stink bug has appeared in increasing numbers this fall in upper New York and other mid-Atlantic states, they have not yet shown their crop-eating presence to any noticeable degree here, according to Connecticut Agricultural Station entomologist Dr. Chris Maier.



The brown marmorated stink bug. Photo by Dr. Chris Maier.

But he said recently that his best guess is they will sharply boost their foul smelling presence in Connecticut within the next two years.

Scientists such as Dr. Maier often play the role of detective, using the known to discover the unknown. Sometimes such detective work involves being a monitor—keeping an eye on a potential problem that, if allowed to develop, could cause harm—and collecting enough data to create practical methods to solve or contain it.

Dr. Maier is doing just that by working to develop a trap or improve an existing trap that will tell farmers and property owners when the brown marmorated stink bug is threatening to infest their fields and properties.

Dr. Maier explained that the stink bug—which won its name because of the foul odor it gives off as a defense mechanism against predators—is a voracious eater of many crops. “They eat flowers, buds, stems, fruits, vegetables—almost anything not made of wood,” he said. “There are effective pesticides that

can kill them but too often stink bugs are not detected in a field until it’s too late and the damage is done.”

He stressed that not enough of them have been found in Connecticut to mount a serious crop threat yet. “But we hope the trap we are developing will give people proper warning before they do,” he said.

So far homeowners have been the most affected in Connecticut, he said, because stink bugs – which proliferate in the spring and summer – tend to seek shelter in September and October to avoid harsh winter weather. And that often means they enter homes and other buildings.

“They don’t sting,” Maier said, “though if touched they may release their foul odor. But people don’t like bugs crawling around their houses.”

Aside from a few effective pesticides (which should not be used in homes) an imported Asian wasp called *Trissolcus halyomorphae* shows promise for reducing bug numbers in nature, killing an impressive 70 percent of stink bug eggs in one sample taken in China. Native wasps have been far less effective, killing only 2 to 5 percent of the insect’s eggs.

The marmorated stink bug is brown, about five eighths of an inch long, and is distinguished by white bands on each antenna.

The vinegar fly or spotted wing drosophila is another insect being studied. This is an Asian pest like the stink bug that feeds on tree fruits such as peaches, plums, cherries and small fruits including raspberries, strawberries and blue berries. It can destroy crops if not discovered early on.

Station scientist and entomologist Richard Cowles is working on monitoring these insects to alert fruit growers of their presence in order to protect their crops. He is working on



The spotted wing drosophila. Photo by Dr. Richard Cowles.

developing an attractive insecticidal bait that can be placed outside crop areas, literally pulling the flies away from the areas where they do damage, then inducing them to feed on an insecticide that will kill them.

The brown marmorated stink bug and the spotted wing drosophila are only two of hundreds of insects that threaten fruit, vegetable and other food crops. Station scientists have been working for years to develop safe, non-chemical means to protect crops and prevent infestations—or to find ways of using effective pesticides for brief periods or “windows” so they will not be harmful in the long run.

Former Station Director Louis Magnarelli has called this anti-destructive bug program, Integrated Pest Management (IPM). He said, “A successful IPM program for these pests will result in minimal cost to growers, less insecticide residue on fruits and preservation of beneficial insects and wildlife found on farms.”

Recent Work by Station Scientists

ARTICLES PUBLISHED

Dr. De-Wei Li, working with a group from the Yale School of Public Health, found that children with asthma who were exposed to indoor molds such as *Penicillium* and other common allergens significantly increased the risk of wheezing. The results will help parents of asthmatic children to better manage asthma. (Environmental Research 118: 86-93).

Dr. James LaMondia developed a new Connecticut broadleaf cigar wrapper tobacco (*Nicotiana tabacum* L.) ‘B2’, a male-sterile hybrid released by the CT Agricultural Experiment Station resistant to Fusarium wilt, tobacco mosaic virus (TMV), the tobacco cyst nematode (TCN) and blue mold. The hybrid is a result of conventional plant breeding and field and greenhouse selection for resistance. This resistant hybrid allows production in pathogen-infested fields with greatly reduced pesticide inputs, increasing yields, and also reducing environmental and human health risks. (CAES Bulletin 1031).

A new fungal pathogen of boxwood was responsible for millions of dollars of losses in Connecticut. **Dr. James LaMondia** tested the pathogen for ability to cause disease in *Pachysandra* (Japanese spurge) and it was shown to be pathogenic. This was the first report of *C. pseudonaviculatum* causing a leaf spot disease on *Pachysandra terminalis*. (Plant Disease 96: 1069).

Jason C. White and collaborators at the University of Massachusetts and the Ocean University of China have found that copper oxide nanoparticles were significantly more toxic to corn than were bulk copper particles or ions. Electron microscopy confirmed the uptake and translocation of the nanoparticles to the shoot tissues. In addition, a novel process involving transformation and transfer of these toxic particles from shoots to back to the roots was also described. These findings are significant because current regulatory guidelines assume nanoparticles and corresponding bulk elements have equivalent bioavailability and toxicity. (Environmental Science and Technology, April 2012).

Drs. Philip M. Armstrong and Theodore G. Arendreis developed a new multi-target real-time reverse transcriptase PCR technique (TaqMan assay) to improve the molecular detection of eastern equine encephalitis virus (EEEV) in mosquitoes collected in the state-wide surveillance program. They added a second primer/probe set to provide redundant targets for EEEV detection. The new assay had similar performance characteristics to the conventional assay but also detected newly evolving strains of EEEV that were not detectable with the conventional assay. The approach increases the reliability of TaqMan assay by creating backup targets for virus detection without sacrificing sensitivity or specificity. (Vector-Borne and Zoonotic Diseases 12:872-876, 2012).

Dr. Mark June-Wells, Dr. Charles R. Vossbrinck, Jordan Gibbons and Gregory Bugbee found that Connecticut’s aquarium trade can be a source of invasive aquatic plants in lakes and ponds by visiting 70 retailers from 2008 – 2010 and documenting the sale of several banned species that they identified visually and by DNA sequencing. Nearly 30% of stores sold banned aquatic plants including: Fanwort, Brazilian waterweed, Parrot feather, and variable milfoil. Fanwort represented > 50% of the banned species sold, while Brazilian waterweed was often mislabeled and sold in 15% of the stores. 7% of the chain stores sold banned species compared to 56% of independent retailers. All stores were informed of State laws prohibiting the sale of invasive species and supplied with the CAES invasive aquatic plant identification guide. (Lake and Reservoir Management 20:200-205, 2012).

Dr. Joseph J. Pignatello and colleagues from the Department of Chemical and Environmental Engineering at Yale University studied the photolysis of the female hormone, estrogen, in freshwater and seawater using artificial sunlight. Estrogen is discharged in sewage treatment effluent. Photolysis of estrogen occurs in pure water

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Dr. Louis A. Magnarelli 1945 – 2013



Dr. Louis A. Magnarelli lost his courageous battle with a blood disease on July 11, 2013. He was an expert on tick-borne diseases and began working at the Experiment Station in July 1975 and was appointed Director in 2004.

As Director, Dr. Magnarelli oversaw the building of the Jenkins-Waggoner Laboratory and established the Grisworld Research Center. He was an effective advocate for the Station and loved and respected by us all.

Dr. John Anderson, Director Emeritus, speaks for all of us when he says, “I shall miss this humble superb scientist, friend, and colleague.”