Frontiers of Plant Science

A REPORT FROM THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION, NEW HAVEN

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The fruit of our labor

By Roger B. Swain Science Editor *Horticulture* Host, *Victory Garden*

It is a great honor to be here to deliver the Samuel W. Johnson Memorial Lecture. I should begin by thanking all of you for the chance to come and speak on this glorious day and to join in this glorious celebration. I do have a day job. It's as science editor of *Horticulture* magazine. For those of you who don't already subscribe, this is what the magazine looks like. You'll also find my picture in the New England magazine called, *People, Places, and Plants*. For those of you who are missing me on the new *Victory Garden*, you'll soon see my face on a new television show called *People, Places, Plants: The Gardening Show*, which is scheduled to begin filming in the spring of 2003.

This lecture has the title: *The Fruit of Our Labor*, because for me this is the favorite part of the gardening year. It's when after all the work and worry despite late frost and other near disasters, we actually do get a taste of our own crops. In my case, I took a short ladder out in my backyard at my farm in New Hampshire yesterday morning and climbed up into the only tree that had ripe apples on it. I only picked two boxes because that's all the apples that were on the tree. You get the whole harvest. Those of you who haven't been doing too much flying and still possess a jackknife should use it to share your apple with your neighbor because by my count there are about 250 apples in this box and it sure looks to me like there are at least twice that many people in the tent. So share your apples because I want to talk today about gardening that benefits your neighbors.

I'm going to offer six steps toward being good to yourself and good to your neighborhood. I begin by reminding you of *the right way to eat* and that's why we start with this apple. This apple is not a great apple. For starters it's as close to an organic apple as I know how to grow, so those of you who are familiar with apple diseases and apple pests will no doubt recognize a little bit of scab and a little bit of curculio damage on these apples.

The variety is July Red. I've been told it was bred at the New Jersey Agricultural Experiment Station about 50 years ago. It was introduced in the early '60s as a better summer apple. Well, a better summer apple means it goes all the way from C- to C+. This apple is good for about 15 minutes and I think this is probably its time. I had them on ice last night and I got them to you as fast as I could because tomorrow they would otherwise split and fall right off the tree. Nevertheless, this apple is at the moment the apple of my eye. If you'd grown it, it would be the apple of your eye too. Because the fact of the matter, and we all know it, is the food we grow ourselves tastes better than anybody else's. The first rule of eating well is to grow your own. I realize that

Samuel W. Johnson Memorial Lecture delivered at Lockwood Farm, Plant Science Day, August 7, 2002 some of you are living in apartments or condominiums where they don't allow you to garden. So persuade your neighbor to let you garden in his or her yard. Now if you absolutely can't raise your own, second best is buying an apple that was grown right here in Connecticut. And that's not hard, You go to a farmer's market. The lovely lady in white that's passing out the apples is my wife Elisabeth. (I'm not married to the cook on the television show!) And the other person passing out the apples is Dan Perkins. He is the father of the husband of the managing editor of Horticulture magazine. So we have a family operation here. Farmers markets, community supported agriculture (CSA), roadside stands-I don't care where you buy your Connecticut apples, but buy Connecticut apples! And finally, if for some reason you can't grow your own and you can't buy locally, then for heaven's sakes when you get to the grocery store please buy something that still has skin on it. That's what we call whole foods. The fastest growing part of the food market is food that has already been cut and split and cooked and packaged. You buy your salad, tear open the plastic bag, and shake it out on the plate. This is not good. You want to buy food that looks the way it grew and then you want to fix it yourself. Now why am I telling you about how to eat. Not only is it good for you, better tasting, it's better for your health. It's also good for your community, because when you raise it yourself you're out there tending the land yourself. When you buy it locally produced, you're supporting local farmers. And when you're buying whole food, you're saying to somebody that you care about what you eat.

You know those supermarket scanners. Is anybody besides me bothered by having everything I buy run through a scanner? There is a computer collecting the data; they're keeping track of what I'm eating. I'm like a dog, I don't like people looking over my shoulder while I eat. But, I've recently become convinced that it is useful to think of the scanner as a voting booth. When we buy something in a supermarket that's whole and we run it through that scanner, somebody records that fact. If the produce has come from far away the purchase may not be good for our immediate neighborhood, but at least it's good for somebody's neighborhood.

For point number two, I've chosen the title *The Seeds are* not the Pits. When you buy a watermelon that has seeds in it, it's half the price for starters. Who thought watermelons shouldn't have seeds. For that matter, what's all this talk about seedless grapes? I raise 25 kinds of grapes. (I hope you go out and take a look at the vineyard just outside this tent). Of these 25 kinds of grapes, I have one vine each and almost all of them have seeds. People bite into that grape and say, "Oh this is one that has a seed." You're allowed to spit out the seed, you're allowed to swallow the seed, but you're not allowed to complain about the seed. The plant makes a big, fleshy, sweet fruit so you will be persuaded to eat it and dump the seeds somewhere else. That's the deal. You ignore the seed and you're breaking the contract. And it's ok to swallow your seeds. Seeds are good, even apple seeds—just don't eat a whole cupful. Virtually all the diversity in grapes, 99.9% of all the diversity in grapes, comes from seeds. Yes, there are some vegetative mutations, but seeds are the primary source of variation.

I have a row of black walnut trees that I raised from seeds that I was given when I visited the Gettysburg battlefield. They are walnuts from a so-called "witness tree" that was standing when the Civil War battle was fought. I'm a whole lot prouder of those black walnut trees than if I had gone to the nursery and bought a plant that somebody had just grown and sold to me. As any horticulturist will tell you, if you want to really understand a plant raise it from seed.

Yes, it's time consuming waiting for a seedling to mature, which brings me to my third point, this enthusiasm for low *maintenance gardening*. Everybody thinks low maintenance gardening is good. Surely it frees you up. Ha! You end up spending more time driving the kids around. I don't know what it's freeing you up to do; but ladies and gentlemen we have to be careful about low maintenance gardening. Low maintenance gardening carried to its extreme is no garden at all. Or as I title this point, "How Low Is No?" There comes a point when you're not there at all and the garden is taking care of itself. A garden taking care of itself is not a garden. It's not a garden unless you have people in it. There are a lot of reasons why you want to be out there. For starters the gardener's shadow is the best fertilizer. Timely intervention is the backbone of integrated pest management. But being out there makes a difference for your neighborhood, too. When someone else comes along and says, "what is that?" you tell them. You talk about the weather, you talk about the neighborhood. It's a point of social exchange. If I were giving a low maintenance lecture, I could have mailed you a cassette to play and we wouldn't have had any of the conversations that I've enjoyed since I got here. And I would have been the poorer for it. Low maintenance gardening has its place. If I was charged with landscaping an office tower, I'd probably use low maintenance gardening techniques. But when I'm working in my own yard, I like the work. I'm not alone. Some people have almost a religious zeal about gardening. Consider me one of them for this is the single greatest skill that human beings could ever come up with.

This brings me to the new Satan, inground irrigation systems. It's wonderful having this podium ladies and gentleman, I get to preach. Inground irrigation systems, first of all, are automated. They pop up to water the lawn when you're not there. Have you ever tried to have a conversation with a pop up sprinkler? It goes *chi-chi-chi* and it wets your pants; *chi-chi* and it wets your pants. This is a really boring conversation. But if somebody has to come out and move the sprinkler, then there's a chance to talk. I don't care what you talk about, but again the social exchange is good for the community. Furthermore, people who water with sprinklers aren't using as much water as people who water with inground irrigation systems. I work in an office that is



Roger B. Swain uses an apple to help make a point..

supposed to be paperless and we fill two dumpsters with waste paper every week. Inground irrigation systems have been found to use more water, not less, by everybody who's measured it. Furthermore, the sprinkler head that has been hit by a UPS truck now sprays straight out into the street.

This is a nice day. Unfortunately we're having too many nice days in a row, which means you're dry here in Connecticut. We're dry in New Hampshire. Indeed the whole nation is conscious that we have to be a lot smarter about water, which brings me to my fourth point-"Well Watered". Some of us have wells. Mine is a dug well and I had to haul water in 5-gallon containers for 4 months last winter because a well went dry. Benjamin Franklin, my ancestor, said 'you only appreciate the value of water, when the well goes dry'. A lot of us are beginning to appreciate the value of water, so we have to use the water more intelligently. Your lawn does NOT need water. I'll give \$100.00 to anybody that could show me an established lawn that was killed by drought east of the Mississippi. My wallet is safe. Yes, the lawn may get brown and crispy. Mine was going crunch, crunch, as I walked across it this morning. But there are a lot of droughtresistant plants that will flourish without supplemental water and we should be growing more of them, and save our precious water for what really needs it.

Anybody tasted his apple yet? I know what happens when you try to eat on television. It makes horrible, juicy sounds and then you choke and somebody bangs you on the back, and you have to film the whole thing over again. Now, my last two points are connected. I know one of you is going to ask me what I think about *biotechnology and genetic engineering*. I've just told you that I am a fan of seeds and conventional breeding. Biotechnology's got me worried, its got me worried because I think it's beginning to smell like a multiflora rose.

When I was at Audubon camp in the 1950's we were told that the multiflora rose would prevent accidents. Planted

alongside highways as a living fence it would prevent cars from crashing. They'd drive right into the rose bush and get stuck. There'd be no damage to the occupants or the car. Well we all have way too many multiflora roses now. Some 50% of our food supply now has genetically engineered food in it. The problem is we don't know where it is. I want to draw your attention to a new generation of crop manipulation that has been dubbed pharmcrops. These are conventional crops like corn that have genes put in them for the synthesis of drugs such as heparin. Manufacturing heparin by growing corn would be a very cheap way to make heparin. The only problem is once you're growing heparin who keeps track of it? What prevents that corn from being used for cornflakes? You have heard about the Starlink corn fiasco. A relatively safe and benign Bt corn that was not licensed for human use yet managed to penetrate the entire food system. I'm less worried about Bt corn. It's going to wipe out Bt for those of us home gardeners who rely on it because resistance is going to develop. And I'm less worried about Roundupready plants, because after all, the herbicides they were using before they used Roundup were even more dangerous. But I am worried about this newest use of biotechnology.

Large corporations are heavily invested in biotechnology and among the promised benefits are that we will be feeding the Third World. However, you can't feed people who don't have money unless you give them the technology, and I haven't seen much biotechnology given away just yet. Which brings me to my last point, *generosity*.

It's easy being generous when you are a gardener. The two television shows, "Victory Garden" and "This Old House", were for many years produced by the same production company, and those two shows are as far apart as you can get. "This Old House" is all about buying a house, tearing it down, and building a new one. "Victory Garden" is all about buying one iris plant and 3 years later dividing it and giving 15 iris plants to your friends. If you don't have stuff to give away at the end of the season as a gardener you have done something terribly wrong. And if I had to look for the essence of civilization, something that held us all together, giving away stuff from the garden-plants, bouquets, apples would be my pick. Along with it, though, is this sharing of information about plants. I look out at all of you in this tent, at all the scheduled programs ahead, this is about as good as life gets. Thank you very much.

Identifying fungi and determining their characteristics is key to safeguarding plant resources

By Mary E. Palm

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Talk given at Imported Pests and Pathogens: Biology, Dispersal, and Control, A Conference Commemorating the 125th Anniversary of The Connecticut Agricultural Experiment Station, October 10, 2000

Accurate identification is key to preventing the introduction of non-native pathogens into the United States. All plant material entering the United States is subject to inspection by USDA, Animal and Plant Health Inspection Service, Plant Protection and Quarantine (APHIS/PPQ) officers. If a disease is observed, the fungus is identified at the port or is sent to me or Dr. John McKemy (as the PPQ National Mycologists) for final identification. Based on the identification, i.e. the name, a determination is made as to whether the fungus is pathogenic and whether it is present in this country. If the fungus is a pathogen not known to occur in the United States the shipment may require treatment or be rejected, which can be costly to the exporter and the importer. On the other hand, an incorrect identification could result in losses to growers or to the natural environment if a nonnative pathogen were allowed to enter because it was identified as a species already in the country. Systematic studies are essential to provide tools for accurate identifica-

tion and the knowledge necessary for appropriate phytosanitary decisions.

Systematics is the science of discovering, organizing and interpreting biodiversity, accomplished by taxonomic and phylogenetic studies resulting in biologically meaningful classifications of organisms. The word biodiversity is used to mean the discovering and describing of the fungi present in the United States and worldwide, including knowledge of the biology, ecology, pathology, physiology, and other aspects of these fungi. Systematic studies result in accurate scientific names that are the keys to unlocking information about the organism and predicting characteristics about that fungus and related fungi. For example, in risk assessments, even if complete knowledge about a specific plant-inhabiting fungus is lacking, information about biological characteristics of closely related fungi, as determined by phylogenetic studies, can be used to predict characteristics of that fungus.

Safeguarding plant resources by preventing the introduc-

tion of non-native, invasive fungi is a challenge. First, it is difficult to inspect plant material for the presence of fungi because fungi grow within a substrate, producing enzymes and absorbing nutrients from living or dead organic matter. Until they produce sporulation structures, the fungi themselves generally are not visible or identifiable and infected plants often appear healthy. Second, although fungi may be present in healthy or diseased material, it is difficult to predict whether they are pathogenic. Systematic studies of plant-inhabiting fungi provide that information as well as identification tools, which are important steps in overcoming these challenges in order to safeguard plant resources.

Pathogenic fungi have been studied extensively in few parts of the world, and this is especially true for fungi associated with newly-traded crops or non-cultivated plants that are harvested directly from the natural environment for export. It is estimated that less than 10% of the fungi have been discovered and described and few have been studied extensively to determine general biological characters such as pathogenic capabilities and host range, as well as potential for genetic variability. It is therefore difficult to precisely predict the risk of introducing invasive fungi that is posed by the movement of most plant material.

The body of a fungus is a microscopic threadlikestructure that lives and grows within its food source. Fungi invade a substrate mainly through the production of enzymes that allow the fungus to absorb nutrients and grow. Because of this heterotrophic, absorptive mode of nutrition, fungi can occur, often inconspicuously, on or in all living or dead organic matter such as plants, insects, and nematodes.

Fungi survive over time, and distribute themselves to new substrates by producing spores through asexual (mitotic) or sexual (meiotic) reproduction, or both. The asexually produced spores often serve to disseminate the fungus short distances during one disease cycle, as in the case of apple scab caused by *Venturia inaequalis* or wheat stem rust caused by *Puccinia graminis*. These spores are disseminated to susceptible hosts by wind, water, insects, or humans. The sexually-produced spores often serve as survival structures that initiate disease at the beginning of the growing season, e.g. in the spring in temperate regions. Fungi also can survive and be distributed as mycelium within the living or dead host, or as specialized survival structures in the substrate or the soil.

All plants, whether diseased or healthy, are hosts to a variety of fungi. Some fungi such as the rusts (Uredinales), smuts (Ustilaginales), downy mildews (Peronosporales), and powdery mildews (Erysiphales) are obligate parasites and must obtain their nutrition from living plants. These fungi usually infect only one host plant species, group of related plant species, or one host plant genus. Other fungi are saprotrophs and decay already-dead organic material. It is largely because of these fungi that fallen leaves and branches are decayed; without them we would be buried in plant debris! Still other fungi cause diseases of living plants but can survive as saprotrophs.



Mary E. Palm identifying a species of *Ravenelia* visible on the screen in the background.

During the past two decades research on the biology of fungi in woody and herbaceous plants has revealed fungi that do not fit easily into those general biological categories. These fungi, called endophytes, occur within nearly all living plants and are generally defined as fungi that cause no apparent harm to the plant. The distinction between an endophyte that causes no disease and a latent plant pathogen that elicits disease symptoms is not clear.

Applying biologically meaningful scientific names for fungi has become increasingly difficult. Obligate parasites such as the rusts, smuts, powdery mildews, and downy mildews generally are host-specific and this is usually reflected in their scientific names. Plant pathogenic fungi that function as facultative saprophytes also have been considered host specific; many species are described and identified on the basis of their host. Recent molecular studies of genera such as Phomopsis and Phyllosticta have shown that such assumed host-specificity does not have a sound genetic basis. Strains of *Phomopsis* from a single plant host species have been found to be genetically diverse, suggesting that species cannot be defined by their plant host alone. Conversely, strains of Phomopsis that are genetically identical, and therefore should be considered one species, may infect a number of different plant hosts. Because accurate identification is essential for preventing the introduction of non-native pathogens, new methods for differentiating these fungi must be developed along with data on their host specificity.

In general, the introduction of plant-disease causing fungi can be traced to the introduction of the host plant. Such fungi have been introduced on all types of cultivated and non-cultivated plants including annuals, perennials, and herbaceous and woody plants.

Plant-inhabiting fungi often are transported by humans, primarily in association with their propagative or nonpropagative plant hosts. The greatest volume of plant material enters the country commercially, but plant material also is introduced by individuals such as plant hobbyists, tourists bringing back a souvenir, or immigrants who want to



A normal poinsettia plant above left. At the right is a diseased poinsettia plant infected by the fungus *Sphaceloma*, which causes the disease called poinsettiae scab. A pathogen of quarantine significance, it causes premature defoliation and elongation of the stem, making poinsettias unsuitable for sale.

bring something familiar from their home country.

Propagative plant materials, such as annual and perennial plants, seeds, and plant cuttings, pose the greatest risk as a pathway for successfully introducing non-native invasive fungi. This is due to the fact that these substrates are hosts to myriad fungi, many of which may be pathogenic but not detectable by visual inspection. Also, information on the existence of pathogens in particular hosts and their host specificity is incomplete. Because of the difficulties in detecting pathogens associated with propagative plant material and the risk that introduced pathogens pose to cultivated and natural hosts, new and creative methods for preventing the introduction of non-native pathogens must be developed. These could include production of "clean" plants by growing under conditions preventing specific diseases, more direct biodiversity data on pathogens associated with a host in a particular area, and new means for treating plants.

Seeds serve as an efficient pathway for the introduction of many plant diseases. Infected seed often is not visibly affected and measures other than inspection are needed to detect these pathogens in order to prevent the introduction of new ones.

Plant cuttings often are imported for propagation. For example, poinsettias previously propagated mainly in the United States are now propagated in several other countries and cuttings are shipped here for distribution to growers. It is possible that poinsettia powdery mildew was introduced accidentally on cuttings from plants grown outside the United States. Poinsettia scab, caused by *Sphaceloma poinsettiae*, present in Florida but considered a quarantine disease by that state, was recently shipped to growers throughout much of the country on cuttings propagated outside of the United States.

Considerable quantities of non-propagative plant materials are transported throughout the world. Such goods include fruits, vegetables, cut flowers, dried herbs, wood, and wood products. Most of this material is consumed or otherwise destroyed and the associated fungus destroyed at the same time, thereby lowering the risk of introduction of non-native pathogens through this pathway. However, in some cases non-propagative material poses a risk due to placement near susceptible hosts. For example, oak sprays with leaves attached are imported. These sprays frequently are used in funeral arrangements, which may be placed in a cemetery, perhaps beneath stately oak trees. Additionally, importation of white rust-infected chrysanthemums for the fresh cut-flower market poses a risk because of the likelihood that infected material may be shipped to florists with adjacent greenhouses or infected material will be discarded by consumers near chrysanthemums in the landscape.

Raw wood and wood products harbor large numbers of diverse fungi and may be stored and used over a long period of time, often outdoors. It is difficult to predict the risk posed by many of these fungi because some wood-inhabiting fungi, harmless to the trees in their native habitats where the fungus and plant host co-evolved, may cause severe problems when introduced into a new environment. Raw wood and wood products also pose a risk because the majority of the fungi associated with this material cannot be observed through inspection. Adding to this risk is the fact that many of the fungi in wood and wood products are vectored by insects and, once introduced, can move long distances very rapidly, especially if the vector is already present or is introduced at the same time.

For the reasons above, the most likely means of successfully introducing non-native pathogens into a new environment is with their associated plant. The risk posed depends on the biology of the fungus, e.g. survival capabilities, spore dispersal characteristics, and host range; it depends also on the type of plant material that is being moved, especially when considering non-propagative material.

Propagules of fungi, especially those that cause root and wilt diseases, can easily be moved in soil, and for this reason soil is regulated by most countries. Wingfield and his colleagues discussed the likelihood that a serious root disease of conifers caused by *Rhizina undulata* was introduced into South Africa in soil with pine trees that were brought by the Europeans into that region. On the positive side, it is probable that many of the mycorrhizal associates, which help plants establish and survive, were introduced in roots and soil along with their plant host.

Pure cultures of fungi are often exchanged between scientists or purchased from culture collections and are usually used only for laboratory research, especially with the advent of molecular techniques in the past decades. If fungi in pure culture are used in the laboratory under appropriate conditions and disposed properly, this pathway poses little or no risk. On the other hand, if these fungi are to be used in greenhouse or field studies, the risk is substantially greater, and each situation must be evaluated carefully.

Some diseases are spread long distances by natural phenomena. For example, sorghum ergot was a serious disease of sorghum in Africa and Asia for nearly a century. It was not present in the New World until it was observed in Brazil in 1995 and Australia in 1996. Within 2 years it was detected in South America, through Central America and the Caribbean and into the southern United States. This fungus produces large quantities of windborne spores on mature, infected heads of sorghum, and these spores are blown long distances. For this reason stopping the spread of this disease was virtually impossible. However, this example points out the importance of an international focus on monitoring diseases and controlling them before they can move to other parts of the world.

New disease-causing fungi are being discovered regularly. Increased knowledge of the biodiversity of fungi will result in increasingly accurate predictions of risk. This knowledge is obtained through systematics and would include the discovery of novel fungi as well as the study of pathogens already known. Systematic studies also are needed to elucidate fungal breeding systems, genetic variation, potential for genetic recombination, etc. This will provide important information about the risk posed by the reintroduction of fungi that are already present in a region.

Due to the dramatic increase in international travel and trade in the global marketplace and because of concern over recent introductions of invasive organisms such as the Asian long-horned beetle and karnal bunt, APHIS-Plant Protection and Quarantine recognized the need to enhance its safeguarding efforts. The National Plant Board was asked to conduct a review of the current system and the resultant "Safeguarding American Plant Resources" provided a thorough analysis and made more than 300 specific recommendations on how to expand and enhance current activities to effectively protect American agriculture and plant resources from entry and establishment of new invasive plant pests and pathogens in the 21st Century. Risk-based management was an overriding theme in the review. Determination of risk relies on scientific information, and international treaties and regulations require that phytosanitary decisions and regulations be based on science.

Pest risk assessments are an essential part of the safeguarding process. These assessments are used to estimate the likelihood that an organism will arrive, survive and thrive in a country. They are the initial step in the decision-making process when a country, industry, or individual requests the opportunity to bring a plant or plant product to the United States. The precision of the risk assessment depends directly on knowledge of the systematics and biology of potential pathogens. The more information available, the more precise the risk assessment will be, and the better plant resources can be successfully safeguarded from invasive fungi. Increased knowledge of the biodiversity of fungi based on systematic studies is essential for providing the scientific information used in such risk assessments.

In conclusion, we have increased knowledge of the systematics and biology of plant-associated fungi, and molecular tools are providing the means for identifying fungi and determining fungal relationships at many levels. When the molecular studies incorporate morphological data the results increase the ability to accurately identify taxa. These tools have increased our ability to track the movement of fungi to help understand how they move from one country to another. They also help in determining the risk posed by reintroducing pathogens already present in a country due to the increased genetic diversity of that pathogen which could result in an increase in pathogenicity or host range or both due to genetic recombination or hybridization of species. Systematic studies using all available tools will increase precision of risk assessments in support of scientifically sound plant quarantine decisions and policies that support world trade of agricultural commodities while protecting the plant resources of individual countries.

West Nile virus found in 15 towns during 2002

Since the fall 1999 discovery of West Nile virus-infected mosquitoes in Connecticut, the virus has been detected in 43 states. Through October 23 nationally there have been 3,296 human cases with 182 of these cases fatal.

During the 2002, crews from the Experiment Station began setting out traps on June 1 at 91 locations in 72 municipalities throughout the state every 10 days. In addition 52 supplemental sites were used for various periods. The mosquitoes were transported to the laboratory each morning, identified, and pooled (grouped) according to species, collecting site, and date. A maximum of 50 female mosquitoes was included in each pool.

Virus isolates from each pool were tested for the following viruses in addition to West Nile: eastern equine encephalitis, Jamestown Canyon, Cache Valley, Highlands J, LaCrosse, and St. Louis encephalitis. In 2002, human cases totaled 17, with cases in Meriden, Greenwich (3), Plainville, Stratford, Hartford (3), North Haven, Bridgeport (2), Ansonia, Stamford (2), and West Hartford (2). In addition, infected horses were reported in Canterbury, Canaan, and Wallingford.

As of October 21, a total of 179,382 mosquitoes had been collected and identified and 15,655 pools of 50 or less female mosquitoes of the same species from the same site had been tested. A total of 305 pools were positive for WNV.

The first WNV-infected mosquitoes were detected on July 24, two weeks prior to onset of the first human case in Meriden. The number of positive pools increased steadily through July and August and peaked during the first week of September. The peak in September coincided with the peak number of human cases.

WNV isolations were from six species of mosquito collected in the following 15 towns: Bridgeport, Darien, Easton, East Haven, Greenwich, Hamden, Hartford, Manchester, New Britain, New Haven, Newington, Norwalk, Shelton, Stamford, and Stratford.

WNV was isolated from 272 positive pools of *Culex pipiens* (principally a bird-biting species) at 20 sites. Two other *Culex* species, *Culex salinarius* and *Culex restuans*, accounted for 24 additional positive pools. WNV was detected in four pools of *Aedes vexans* from three sites, *tw pools of Ochlerotatus trivittatus* from two sites, and *Aedes cincereus, Ochlerotatus sollicitans, and Uranotaenia sapphirina* in one pool each.

The basic conclusions that can be drawn include the following: There is continued activity with no sign of decline, that *Culex* mosquitoes seem to be the most important vectors, that virus isolations from mosquitoes are consistent with bird mortality and human cases in time and space, that *Culex salinarius* and *Aedes vexans* are the most suspect vectors to humans and horses, and that reemergence of WNV can be anticipated again next year.

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