ECOLOGY OF EMERGING VECTOR-BORNE PLANT DISEASES

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Individuals, populations, communities, and ecosystems are impacted by pathogenic organisms at different levels. During epidemics, high mortality may result in temporary or permanent perturbations of ecological networks within communities (Daszak et al., 2000). Alterations in community structure can significantly impact habitats driving ecosystem change. On the other hand, anthropogenic environmental changes may have catastrophic consequences to natural communities and populations, in some cases resulting in pathogen spill over to humans (e.g., Daszak et al., 2000; Patz et al., 2004; Power and Mitchell, 2004). Therefore, reducing the social, economic, and environmental impacts of diseases requires in-depth knowledge of pathogen, host, and vector biology and ecology.

The increasing number of emerging diseases and epidemics in recent decades has stimulated interest in understanding how new diseases arise and previously rare diseases increase in incidence. However, most of the research linking diseases with environmental change has been limited to human and animal pathogens (e.g., Daszak et al., 2000; Patz et al., 2004). In this essay I will argue that there are important connections and similarities between human diseases and plant diseases, focusing on those occurring in agricultural systems. I will also discuss similarities among vector-borne diseases and present an example of how the introduction of an invasive vector species has dramatically modified the ecology of a bacterial pathogen of previous limited importance. One of my main goals is to emphasize that much could be gained in our understanding of the ecology of vector-borne human and animal diseases from work done with agricultural systems, and vice versa. Unfortunately, to this date researchers in these two domains remain largely unaware of each other.

Emerging Vector-Borne Diseases

The number of disease epidemics has dramatically increased in recent years, as have the threat of emerging new diseases and the reemergence of other diseases. Although biological factors such as pathogen mutations have been demonstrated to be associated with recent epidemics (Anishchenko et al., 2006), surveys have suggested that most diseases can be linked to anthropogenic activities (Woolhouse and Gowtage-Sequeria, 2005). A growing body of literature exists on pathogens disseminated without the aid of vectors, such as primate viruses "jumping" to human hosts primarily due to bush meat hunting activities (Wolfe et al., 2005).

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Commerce, frequency and speed of transportation, invasive species, pesticide resistance, urbanization, climate change, and many other factors have been linked to emerging human diseases (Woolhouse and Gowtage-Sequeria, 2005). Anderson et al. (2004) conducted the only survey that systematically studied factors driving emerging plant diseases. Although introductions (56 percent) and weather (25 percent) were determined to be responsible for most emerging plant diseases, other factors were also found to be of importance. Interestingly, it was observed that viruses composed 47 percent of all emerging plant diseases. A similar trend was found in human emerging diseases (Woolhouse and Gowtage-Sequeria, 2005). Therefore, emerging human and plant diseases share driving factors, and approaches to control either one might be instructive to researchers working with both groups of pathogens.

Human Health, Environmental Change, and Plant Diseases

One of the challenges for this century will be to sustainably produce enough food for an exponentially growing world population. In 2006, 6.5 billion people inhabited the planet; the World Health Organization estimates that number will increase to 9 billion by 2050 (UN, 2007). Increasing crop yield with sustainable agricultural approaches that are not detrimental to the environment will be challenging. Approximately 40 percent of the world's yield is currently lost due to pests (pre- and post-harvest) (Agrios, 1997). Because malnutrition and poverty are directly linked to human health, sustainably producing increased quantities of food to populations around the world will be a global challenge for future generations.

The increased technological inputs for agriculture and the expanding scale of monocultures provide continual change in challenges for producing food. The expansion of agricultural land and increased pesticide, irrigation, and fertilizer use have been the major controllable inputs to increase crop yield. These alternatives have various detrimental effects on the environment and human and animal health. Furthermore, the long-term sustainability of these strategies is questionable due to the environmental impact of current agricultural technology (Altieri and Nichols, 2005). To increase farm land one must explore new regions, infiltrating into forest, grassland, or other habitats that may provide important ecosystem services. An increased human-natural vegetation interface may also result in new human and plant diseases, as pathogens may spill over from natural environments into new host organisms (Power and Mitchell, 2004). To reduce losses and increase yield per unit area, pesticides and fertilizers must be applied in increasing quantities. The environmental and health impacts of pesticides have been highly publicized. Fertilizers have a similar reputation, for example their role in driving toxic algal blooms caused by agricultural runoff in waters throughout the world (Gilbert et al., 2006). Because food production is tightly connected to human health, promoting sustainable agricultural practices

may reduce the impact of human pathogens from individual to population levels. Quantitatively determining the importance of plant health in the maintenance of a healthy human population and a sustainable environment would certainly be an interesting exercise.

Contrasting Plant and Animal Vector-Borne Diseases

A large diversity of organisms transmits plant pathogens. The most common vectors are insects, but mites, nematodes, and fungi are also important (Agrios, 1997). Insects transmit plant-pathogenic viruses, bacteria, fungi, nematodes, and protozoa. Among insects, sap-sucking hemipterans such as aphids, leafhoppers, planthoppers, and whiteflies are the major vectors. Of those, aphids are the most important group, as they are responsible for disseminating 70 percent of vector-borne plant viruses (Nault, 1997). Like vector-borne animal pathogens, vector-plant pathogen interactions can be classified based on several characteristics, such as requirement for circulation and/or propagation within vector and temporal characteristics of transmission and pathogen retention (Gray and Banerjee, 1999; Ng and Perry, 2004). Molecular determinants of vector transmission have been well explored for only a few plant disease systems compared to numerous animal disease systems (e.g., Gray and Gildow, 2003).

In addition to commonalities in transmission biology, the ecology of vector-borne diseases of plant and humans also share important similarities. Human vector-borne pathogens are generally categorized as the etiological agents of anthroponotic (human-centered) diseases such as malaria or zoonotic (having an animal reservoir) diseases such as Lyme disease, in relation to their ecology (Eldrige and Edman, 2000). This is an important distinction with epidemiological implications, as the involvement of animal hosts in addition to humans in zoonotic diseases must be well understood to devise control strategies to reduce pathogen spread. A similar scenario occurs with plant pathogens. Some insect vectors are host-specific (e.g., certain aphid species), whereas others can have broad host ranges (e.g., sharpshooter leafhoppers). The host range of plant pathogens is largely dependent on the degree of vector specificity required for efficient dissemination and on the host range of the vectors, as usually there are no other means of spread. Pathogens transmitted by species with narrow host ranges tend to be plant specific, whereas those transmitted by polyphagous insects may infect many plants and cause disease in several crops or weeds. Some phloem-limited bacteria (mollicutes) of maize, for instance, only colonize species in the plant genus *Zea* (maize and teosintes) and are spread by a few oligophagous leafhopper vectors that have co-evolved with those host plants (Nault, 1990). In contrast, insect transmission of the bacterium *Xylella fastidiosa* has low vector specificity, being transmitted by several sharpshooter leafhoppers and the more distantly related spittlebugs. Both of these insect groups tend to be polyphagous (Redak et al., 2004). In consequence, this pathogen could colonize hosts in at

least 94 species tested in 28 different plant families (Hill and Purcell, 1995a). Therefore, plant pathogens with a very narrow host range behave ecologically as anthroponotic diseases, whereas those with a wide host range behave more similarly to zoonotic ones.

Nevertheless, several relevant differences must be kept in mind when extrapolating concepts from animal to plant systems or vice versa. Host movement is significantly different in these systems. The host immune response of plants is very different from that of animals. Host genetic diversity may be high in animal systems, but is usually extremely low in crops. Moreover, the spatial, age structure, and population densities of crop plants differ dramatically from those of animals. In addition, plant disease epidemics often are not categorized as such unless at least thousands of individuals are infected. Therefore, host social networks, movement, immune response, and recovery are not considered of importance in plant epidemiology. Conversely, other approaches that incorporate the availability of large numbers of static susceptible hosts are more useful for plant systems.

The Plant Pathogenic Bacterium *Xylella fastidiosa* as a Case Study

The xylem-limited bacterium *X. fastidiosa* is present throughout the Americas and causes disease in many crops of economic importance, including Pierce's disease of grapevines (PD), almond leaf scorch (ALS), and citrus variegated chlorosis (CVC) (Purcell, 1997). *X. fastidiosa* is disseminated among plants by sharpshooter leafhoppers (Hemiptera: *Cicadellidae*) and spittlebugs (Hemiptera: *Cercopidae*), both of which specialize in feeding on the sap in plant xylem (water-conducting tissue) (Severin, 1949, 1950). Sharpshooter leafhoppers are considered the most important vectors in epidemics examined so far. Transmission is not specific, as different strains of *X. fastidiosa* are transmitted by different vector species. There is no transmission of *X. fastidiosa* from parent to offspring and no required latent period (Freitag, 1951; Purcell and Finlay, 1979). However, the bacterium multiplies in the foregut of vectors and is persistent in adult insects but is lost when immature insects molt (Hill and Purcell, 1995b; Purcell and Finlay, 1979). The inoculum of *X. fastidiosa* for plant inoculation is located in the canals leading to the sucking pump (cibarium) of the foregut of vectors (Almeida and Purcell, 2006; Purcell et al., 1979). Transmission efficiency, however, varies dramatically depending on the combination of host plant, bacterial strain, and vector species. The factor most clearly associated with transmission efficiency is bacterial densities within plants, with higher cell numbers resulting in increased transmission rates (Hill and Purcell, 1997). The ecology of *X. fastidiosa* shares similarities with complex zoonotic diseases with multiple host species. *X. fastidiosa* has a very wide host range (Hill and Purcell, 1995a), with colonization patterns varying from systemic pathogenic plant-strain associations to infections that die out over time (Purcell and Saunders, 1999). The host range of sharpshooter vectors can also be very large, with up to a few hundred plants

listed for certain species (Redak et al., 2004). Because *X. fastidiosa* has such a wide host range and is vectored without specificity by a group of insects that tends to be polyphagous, the resulting diseases have complex epidemiology.

Although *X. fastidiosa* has been present in California for over 100 years, only three large epidemics have occurred in that period of time, all of which were associated with grapevine hosts (PD) (reviewed by Hopkins and Purcell, 2002). The first one occurred in the late 1800s in Southern California, which decimated the incipient grape industry in the region. In the 1930s to 1940s an epidemic in the Central Valley associated with infected sharpshooters migrating from alfalfa fields was also of importance and resulted in several breakthroughs in our understanding of *X. fastidiosa* diseases by researchers at the time. In recent decades, however, the disease has been constantly present at low incidence in the wine grape growing coastal valleys of Napa and Sonoma. The third, and current, epidemic emerged after the introduction of a polyphagous invasive vector species, *Homalodisca vitripennis* (glassy-winged sharpshooter; Hemiptera: *Cicadellidae*) (Sorensen and Gill, 1996), into Southern California in 1989 (Blua et al., 1999). This invasive species is the driving factor of PD epidemics in Southern California and the southernmost region of the Central Valley. It is also responsible for several emerging *X. fastidiosa* diseases in California, such as oleander leaf scorch. I will discuss the current hypothesis on how *H. vitripennis* has increased the incidence of PD and how it may be responsible for the emergence of new diseases.

An Invasive Vector Driving the Emergence of a Rare Disease

PD epidemics have occurred in different regions of California, although much of Southern California and the Central Valley have been largely disease free in the last decades. The introduction of *H. vitripennis* into the state dramatically changed this scenario. In 1999, reports of PD outbreaks in the small wine region of Temecula Valley resulted in very high infection rates in just a few years after the epidemic began (Purcell and Feil, 2001); a similar situation occurred in Kern County, the southernmost area of the Central Valley, starting in 2000 (Hopkins and Purcell, 2002). A large area-wide monitoring, control, and research project is in place to address this problem and temporarily limit the distribution of *H. vitripennis*. The driving factor associated with the outbreak was the presence of extremely large numbers of *H. vitripennis* in vineyards. This vector overwinters in large number on citrus, up to thousands per plant, and has a larger dispersal range than that of typical sharpshooters. Therefore, it has been suggested that sheer numbers of an invasive species, not under biological control by native parasitoids, predators, or parasites, was the main factor driving the epidemic. Two cycles of pathogen spread could occur in this scenario, one of primary spread by infective vectors migrating from citrus to grape in early spring, and a second cycle with a new generation of vectors on grape that could acquire the pathogen from plants infected earlier in the year and transmit it to new plants during the

summer and fall. Because citrus does not serve as a host of *X. fastidiosa* strains causing disease in grape in the United States, it has been suggested that secondary spread is responsible for the outbreak, a hypothesis dubbed "vine-to-vine spread" (Hopkins and Purcell, 2002).

Although the ecological factors responsible for these outbreaks are not well understood, it is clear that large vector populations are an important component of this system. *H. vitripennis* is a poor vector of *X. fastidiosa* to grape when compared to other species (Almeida and Purcell, 2003). Therefore, its ecology and behavior seem to offset low transmission rates. In addition, *H. vitripennis* can infect dormant vines under field conditions, opening a new window of time for new infections, when infective insects on citrus may migrate to vines in warm days during the winter (Almeida et al., 2005). That may be important because *H. vitripennis* overwinters on citrus and moves to vines in early spring when young shoots are present, remaining in vineyards until the winter (Park et al., 2006). Furthermore, *H. vitripennis* can also inoculate the woody tissue of vines, which are closer to tissues of the plant that are not pruned off during the winter, possibly resulting in a larger number of infections late in the growing season that persist through to the next year (Almeida and Purcell, 2003). On the other hand, evidence demonstrated that some late infections recover by a yet to be determined plant physiological mechanism during dormancy (Feil et al., 2003). In summary, PD epidemics in the presence of *H. vitripennis* in California seem to be driven primarily by an invasive vector species that compensates for poor transmission efficiency by having large populations in and near citrus, and behavioral and ecological characteristics that promote pathogen spread within vineyards.

Emergence of New X. fastidiosa *Diseases*

There are many strains, or genetic clusters, of *X. fastidiosa* isolates (Schuenzel et al., 2005). Like other bacterial pathogens, the difficulty in defining species boundaries, or what a bacterial species is, has plagued the taxonomy of *X. fastidiosa*. Nevertheless, this is a pathogen primarily limited to the Americas, with only one exception in Asia (pear disease) and a report from Europe (Purcell, 1997). Diversity studies have focused on diseased crops, biasing sample collection towards pathogenic isolates occurring in a limited number of host plants (e.g., Hendson et al., 2001; Schuenzel et al., 2005). Pathogenicity studies, linking genetic diversity to host species susceptibility, have not been widely conducted, limiting the interpretation of molecular diversity results. Studies in the United States have provided an idea of *X. fastidiosa*'s diversity, primarily because it causes disease in many host plants in the country compared to Brazil, for example, where it is documented to cause disease in only three crops. If environmental samples from alternative, nonsymptomatic hosts were included in such surveys, it is reasonable to assume that a much larger number of genetic clusters could be identified. As previously mentioned, this is a pathogen transmitted by several

polyphagous sharpshooters with very wide plant host ranges. Thus, this pathogen has the potential to diverge and maybe have a high rate of genetic recombination among isolates in different environments and host plants.

Oleander leaf scorch (OLS) emerged in the mid-1990s in Southern California, and was tightly associated with the presence of *H. vitripennis* (Purcell et al., 1999). OLS is caused by what at the time was a new strain of *X. fastidiosa*. It is possible that this strain of *X. fastidiosa* was present in alternative host plants in the region, with limited dispersal by native vector species occurring in low numbers and without feeding preference for oleander; an alternative hypothesis is that this strain was an introduction into California. It can be hypothesized that *H. vitripennis*, present in high numbers and with feeding preference for oleander, could have acquired this strain from an alternative host and transferred it to oleander where it was maintained by the presence of susceptible hosts by a vector occurring in high number on the same host. A similar mechanism may be responsible for the emergence of *X. fastidiosa* diseases in many host plants in the presence of *H. vitripennis* in recent years, including mulberry, sweet gum, and olive (Wong et al., 2006).

A model for the emergence of new diseases after the introduction of a new vector into a region could be valid for vector-borne diseases in which pathogens are maintained in the environment in hosts of marginal epidemiological importance by vector species with little or no preference for feeding on humans or animals of interest. In this situation, pathogens have the opportunity to not only be maintained in endemic cycles, but also diverge and evolve into new strains, as different vector species may have associations with hosts of variable degrees of specificity. The introduction of a new vector species may result in pathogen acquisition from such cycles and its transfer to new disease cycles where it may be self-maintained (Figure 1-12).

Concluding Remarks

Human, animal, and plant vector-borne pathogens share several biological, ecological, and epidemiological similarities, but important differences exist. Unfortunately, scientists studying these systems rarely exchange ideas or are aware of each other's research contributions. Plant scientists, for example, could incorporate tools and concepts from studies on human diseases that integrate pathogen spatial and temporal distribution and molecular population genetics to develop disease spread and evolution models. On the other hand, plant systems allow large experiments to be conducted, with multiple hosts, vector species, and pathogen strains, which could be used to experimentally address ecological and evolutionary hypotheses on pathogen range and transmission efficiency. Finally, ecological hypotheses based on either system may be useful in building models that can be tested for the development of disease control strategies.

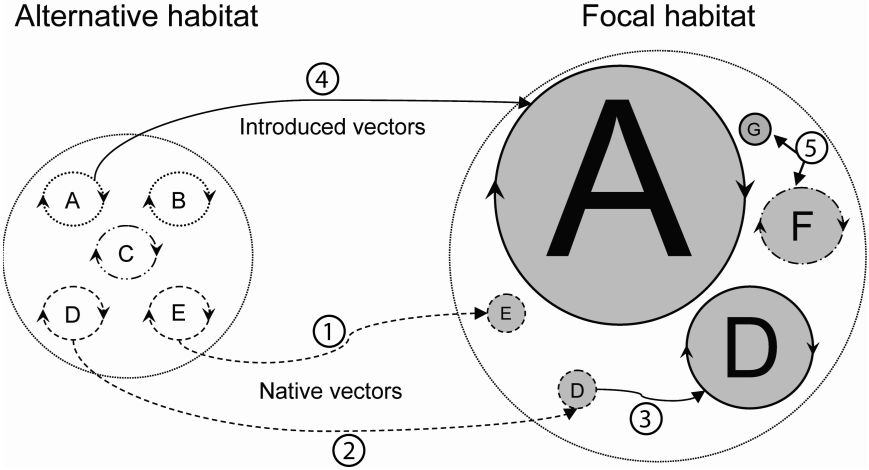

FIGURE 1-12 Model illustrating a hypothesis on how newly introduced vectors may drive new disease epidemics. On the left, different strains of a pathogen (labeled with different letters) are maintained in endemic disease cycles in alternative habitats by different vector species (different dashed-line circle borders). An introduced vector species is indicated by solid lines. On the right, shaded disease cycles occur on hosts of interest (e.g., humans, animals, plants), with circle size representing the dimensions of the epidemic. In this scenario, several pathogen strains are kept in alternative hosts indefinitely by native vectors, with occasional infection of a host of interest with epidemiological consequences (dead-end hosts; spread events [numbered circles] 1 and 2). However, an invasive vector could acquire the pathogen from otherwise dead-end hosts and establish a new epidemic disease cycle (event 3). An alternative may occur when introduced vectors acquire a pathogen from an endemic cycle and establish a new epidemic cycle in a new host, as illustrated with isolate A (event 4). An invasive vector may also introduce novel pathogens (event 5) to focal habitat which may (F) or may not (G) spread. It may also be possible for vectors to move pathogens from epidemic cycles to endemic ones, which would function as pathogen reservoirs for future epidemics.

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