5-2016

Application of genetics and genomics to wildlife epidemiology

Julie A. Blanchong
Iowa State University, julieb@iastate.edu

Stacie J. Robinson
U.S. Department of Commerce

Michael D. Samuel
U.S. Geological Survey

Jeffrey T. Foster
University of New Hampshire, Durham

Application of genetics and genomics to wildlife epidemiology

Abstract
Wildlife diseases can have significant impacts on wildlife conservation and management. Many of the pathogens that affect wildlife also have important implications for domestic animal and human health. However, management interventions to prevent or control wildlife disease are hampered by uncertainties about the complex interactions between pathogens and free-ranging wildlife. We often lack crucial knowledge about host ecology, pathogen characteristics, and host–pathogen dynamics. The purpose of this review is to familiarize wildlife biologists and managers with the application of genetic and genomic methodologies for investigating pathogen and host biology to better understand and manage wildlife diseases. The genesis of this review was a symposium at the 2013 annual Wildlife Society Conference. We reviewed the scientific literature and used our personal experiences to identify studies that illustrate the application of genetic and genomic methods to advance our understanding of wildlife epidemiology, focusing on recent research, new techniques, and innovative approaches. Using examples from a variety of pathogen types and a broad array of vertebrate taxa, we describe how genetics and genomics can provide tools to detect and characterize pathogens, uncover routes of disease transmission and spread, shed light on the ways that disease susceptibility is influenced by both host and pathogen attributes, and elucidate the impacts of disease on wildlife populations. Genetic and increasingly genomic methodologies will continue to contribute important insights into pathogen and host biology that will aid efforts to assess and mitigate the impacts of wildlife diseases on global health and conservation of biodiversity.

Keywords
epidemiology, genetics, genomics, pathogen, transmission, virulence, wildlife disease

Comments
This article is published as Blanchong, Julie A., Stacie J. Robinson, Michael D. Samuel, and Jeffrey T. Foster. "Application of genetics and genomics to wildlife epidemiology." The Journal of Wildlife Management 80, no. 4 (2016): 593-608. doi: 10.1002/jwmg.1064. Posted with permission.

The Journal of Wildlife Management 80(4):593–608; 2016; DOI: 10.1002/jwmg.1064

Invited Paper

Application of Genetics and Genomics to Wildlife Epidemiology

JULIE A. BLANCHONG,[1] *Department of Natural Resource Ecology and Management, Iowa State University, 339 Science II, Ames, IA 50011, USA*

STACIE J. ROBINSON, *National Oceanic and Atmospheric Administration, Honolulu, HI 96818, USA*

MICHAEL D. SAMUEL, *U.S. Geological Survey, Wisconsin Cooperative Wildlife Research Unit, University of Wisconsin, 204 Russell Labs, 1630 Linden Dr., Madison, WI 53706, USA*

JEFFREY T. FOSTER, *Department of Molecular, Cellular and Biomedical Sciences, University of New Hampshire, 291 Rudman Hall, Durham, NH 03824, USA*

ABSTRACT Wildlife diseases can have significant impacts on wildlife conservation and management. Many of the pathogens that affect wildlife also have important implications for domestic animal and human health. However, management interventions to prevent or control wildlife disease are hampered by uncertainties about the complex interactions between pathogens and free-ranging wildlife. We often lack crucial knowledge about host ecology, pathogen characteristics, and host–pathogen dynamics. The purpose of this review is to familiarize wildlife biologists and managers with the application of genetic and genomic methodologies for investigating pathogen and host biology to better understand and manage wildlife diseases. The genesis of this review was a symposium at the 2013 annual Wildlife Society Conference. We reviewed the scientific literature and used our personal experiences to identify studies that illustrate the application of genetic and genomic methods to advance our understanding of wildlife epidemiology, focusing on recent research, new techniques, and innovative approaches. Using examples from a variety of pathogen types and a broad array of vertebrate taxa, we describe how genetics and genomics can provide tools to detect and characterize pathogens, uncover routes of disease transmission and spread, shed light on the ways that disease susceptibility is influenced by both host and pathogen attributes, and elucidate the impacts of disease on wildlife populations. Genetic and increasingly genomic methodologies will continue to contribute important insights into pathogen and host biology that will aid efforts to assess and mitigate the impacts of wildlife diseases on global health and conservation of biodiversity. © 2016 The Wildlife Society.

KEY WORDS epidemiology, genetics, genomics, pathogen, transmission, virulence, wildlife disease.

Wildlife diseases have significant ramifications for global health and the conservation of biodiversity (Daszak et al. 2000). For instance, 60% of emerging infectious diseases in humans are zoonotic, most with a potential wildlife reservoir (Jones et al. 2008*a*). In addition, the rate of newly emerging or re-emerging wildlife diseases has increased (Cohen 2000, Dobson and Foufopoulos 2001). High profile outbreaks of zoonotic diseases (e.g., West Nile virus, avian influenza, Ebola) increased public awareness of the connection between wildlife and human health. However, this relationship is not new; diseases associated with wildlife (e.g., rabies, plague) have beleaguered humankind for centuries. Ongoing globalization, habitat fragmentation, and climate change are increasingly important contributors to the increase in disease emergence and the increasing connection between human, livestock, and wildlife health (Daszak et al. 2000, Harvell et al. 2002, Olden et al. 2004).

Wildlife diseases are increasingly recognized for their potential impacts on conservation and biological diversity. Recent wildlife epidemics such as Tasmanian devil (*Sarcophilus harrisii*) facial tumor disease (DFTD; McCallum 2008) and chytridiomycosis (Skerratt et al. 2007) provide clear evidence that emerging diseases can cause population declines, changes in distribution, or even extinction. Other wildlife diseases can have economic or social impacts. White-nose syndrome, caused by a fungus that recently emerged in North America, for example, has been responsible for the deaths of millions of insect-eating bats, and formerly common species such as the little brown bat (*Myotis lucifugus*) have undergone local extirpations (Frick et al. 2010). To protect human and domestic animal health and maintain healthy and abundant wildlife populations, many state and federal natural resource agencies are increasingly faced with mandates to manage wildlife disease (Deem et al. 2001). However, identifying effective management

Received: 28 August 2015; Accepted: 26 February 2016

[1]*E-mail: julieb@iastate.edu*

interventions is hampered by the difficulty in understanding the complex interactions between pathogens and free-ranging wildlife (Joseph et al. 2013; Fig. 1).

The purpose of this review is to familiarize wildlife biologists and managers with the application of genetic and genomic methodologies for investigating pathogen and host biology to better understand and manage wildlife diseases. The genesis of this review was a symposium at the 2013 annual Wildlife Society Conference. We reviewed the scientific literature and used our personal experiences to identify studies that illustrate the application of genetic and genomic methods to advance our understanding of wildlife epidemiology, focusing on recent research, new techniques, and innovative approaches. Using examples from a variety of pathogen types and a broad array of vertebrate taxa, we describe how genetics and genomics can provide insight into the detection and characterization of pathogens, how genetic methods can contribute to understanding disease transmission and spread, how aspects of disease susceptibility are influenced by genetic attributes of both the host and the pathogen, and how genetic approaches can shed light on the impacts of disease on wildlife populations (Table 1). We conclude with a discussion on emerging and future genetic and genomic applications to wildlife epidemiology. We have provided a glossary with definitions of important terminology for those readers less familiar with genetics (Supplemental Material, available online in Supporting Information). Our review is designed primarily to cover the breadth of genetic and genomic approaches used to study wildlife diseases; it is not intended to be comprehensive because of the vastness of disease literature.

PATHOGEN DETECTION AND CHARACTERIZATION

Sudden cases of weak and moribund animals often indicate wildlife disease outbreaks. Alternately, some diseases are discovered through routine wildlife health surveillance. In either event, the first step of an investigation is determining its etiology (i.e., cause of the disease) and with most diseases this means identifying the pathogen. For well-studied

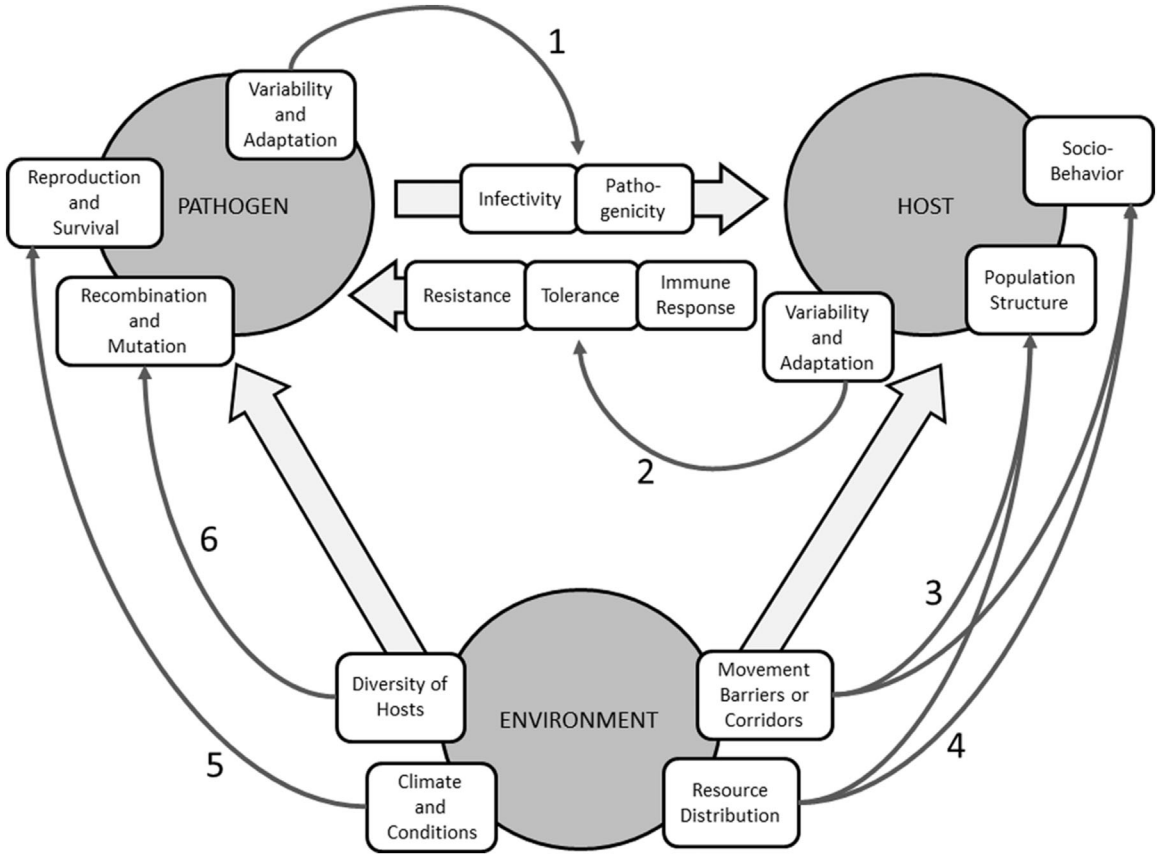

Figure 1. A conceptual diagram of the complex interactions between hosts, pathogens, and their environment that underlie epidemiological processes. Numbered arrows indicate mechanisms by which these interactions affect genetic patterns (of host or pathogen) that may, in-turn, be used to discern crucial information about an infectious disease system. (1) Genetic diversity of pathogens supports adaptation to better infect hosts and optimize pathogenicity; evolutionary signatures detectable in pathogen phylogenies. (2) Genetic diversity of hosts supports adaptation to resist infection, tolerate infection, or mount immune response; evolutionary signatures detectable in host phylogenies. (3) Landscape features affect movement and potential disease spread between populations; landscape genetic analyses of host or pathogen can help identify geographic paths of disease spread. (4) Habitat and resources shape local population structure, host contact, and host–pathogen contact rates; genetic data may inform relatedness or contact network analyses to detect transmission patterns. (5) Environmental conditions affecting pathogen reproduction or survival will affect transmission (either between hosts or from environment to host) and evolutionary potential; phylogenetic or phylodynamic analyses are useful here. (6) Because hosts constitute pathogen habitat, mixing of host species can facilitate reassortment or cross-over events leading to pathogen mutation or host jumps; phylogenetics or phylodynamics are useful here as well.

Table 1. Applications of genetics and genomics to questions in wildlife epidemiology.

Objective	Summary	Examples
Pathogen detection and characterization		
Identify pathogens	Polymerase chain reaction assays can detect and quantify pathogen DNA or RNA in host or environmental samples.	Ranavirus in amphibians (Galli et al. 2006), white-nose syndrome in bats (Lorch et al. 2010).
Characterize pathogens	Whole genome analyses enable greater understanding of mechanisms of transmission, dispersal, and evolution.	*Mycoplasma gallisepticum* in house finches (Delaney et al. 2012), brucellosis in feral swine (Leiser et al. 2013).
Understand disease origin or identify reservoir or spillover hosts	Mutations in DNA or RNA sequences can be used to determine how pathogens from different hosts or locations are related.	Chytrid fungus in amphibians (Rosenblum et al. 2013), *Mycobacterium bovis* in badgers and cattle (Biek et al. 2012).
Disease transmission and spread		
Construct transmission networks	Genetic markers can be used to evaluate the influence of host genealogical relationships on transmission. Temporal or spatial genetic relationships of the pathogen can identify networks that facilitate transmission.	Canine distemper and leptospirosis in raccoons (Dharmarajan et al. 2012), *E. coli* in African ungulates. (VanderWaal et al. 2014).
Explain disease distribution	Spatial scale of genetic correlation among hosts or pathogens can provide insights into the distribution and potential spread of disease.	Bovine tuberculosis in white-tailed deer and elk (Vander Wal et al. 2013), avian influenza in waterfowl (Hill et al. 2012).
Understand how landscape features affect disease spread	Integration of genetic techniques with landscape ecology can identify corridors or barriers to host movement and elucidate how landscape composition affects spread.	Rabies in raccoons (Cullingham et al. 2009) and striped skunks (*Mephitis mephitis;* Paquette et al. 2014), chronic wasting disease in white-tailed deer (Robinson et al. 2013).
Disease susceptibility and pathogen virulence		
Understand how genetic diversity influences disease susceptibility	Heterozygosity or the number of alleles at neutral and functional genes may be associated with probability of infection or probability of survival.	Bovine tuberculosis in wild boar (Acevedo-Whitehouse et al. 2005), Tasmanian devil facial tumor disease (Siddle et al. 2007).
Identify genes associated with resistance or tolerance	Specific alleles at immune or other candidate genes may contribute to understanding an individual's resistance or tolerance to disease.	Avian malaria in house sparrows (Loiseau et al. 2011), chronic wasting disease in cervids (Robinson et al. 2012*c*).
Understand how pathogen virulence and cross immunity affect susceptibility	Genetic variation of pathogens may affect the rate or route of transmission. Interaction with the host immune system can shape pathogen evolution.	Avian influenza in mallards (Latorre-Margalef et al. 2013, 2014), ranavirus in amphibians (Echaubard et al. 2014).
Impacts of disease		
Understand the evolution of resistance	Genetic methods can identify the influence of genetic diversity and disease selection pressure on the development of disease tolerance or resistance.	Chronic wasting disease in white-tailed deer (Robinson et al. 2012*a*), avian malaria in Hawaiian honeycreepers (Foster et al. 2007).
Characterize impacts of disease on wildlife populations	Disease may alter population processes that affect genetic diversity and rates or patterns of gene flow.	Tasmanian devil facial tumor disease (Lachish et al. 2011, Bruniche-Olsen et al. 2013), mange in bobcats (Serieys et al. 2015).

wildlife taxa, classic microbiological techniques and gross pathology can narrow the list of potential pathogens by identifying agents associated with certain pathologies such as tissue damage, while also differentiating pathogens from commensal microbes or incidental environmental contaminants. These traditional techniques include microscopy, growth on various media, cell culture, biochemical tests, immunological (e.g., serology), and other biological assays to identify the causative agent (Silvy 2012). More recently, highly sensitive genetic techniques have been developed to detect pathogens directly from infected animals or the environment. For instance, using sequencing to identify a pathogen from a blood sample or cloacal swab. With adequate validation and quality controls, these genetic approaches can obviate extensive laboratory testing by highly specialized staff.

Genetic detection of pathogens began with simple polymerase chain reaction (PCR) tests for human diseases, with PCR products run on agarose gels to detect molecular weight bands specific for a particular pathogen (Rolfs et al. 1992, Yamamoto 2002). Polymerase chain reaction has now been applied to detection of many wildlife pathogens, including DNA viruses (Galli et al. 2006), bacteria (Bricker 2004), protozoan blood parasites (Hellgren et al. 2004), and fungi (Lorch et al. 2010). Pathogen detection using these PCR approaches is relatively quick and inexpensive and if the assays are designed correctly provide accurate pathogen identification. Recently, real-time quantitative PCR methods, with higher sensitivity and throughput than traditional PCR, have also been applied to a broad range of pathogens, from viruses (Decaro et al. 2005) and bacteria (Roug et al. 2014), to fungi (Boyle et al. 2004, Muller et al. 2013) and parasites (Knowles et al. 2011). Moreover, these assays can often be designed to quantify the pathogen load, which is particularly important when the amount of pathogen is related to the disease state, such as fungal pathogen growth on bats with white-nose syndrome (Langwig et al. 2015). A similar approach using RNA sequence targets, reverse transcriptase PCR (RT-PCR), has been developed for many wildlife viruses such as West Nile virus (Porter et al. 1993), rabies (Black et al. 2002), and avian influenza (Pasick 2008). As with simple PCR, RT-PCR can be used to quantify the amount of viral particles. These

pathogen detection approaches can also be applied to screening disease vectors such as mosquitoes, ticks, and fleas to determine transmission potential (Eldridge and Edman 2004), though these arthropods often need to be pulverized for nucleic acid extraction from within hard exoskeletons (Harrison et al. 2015). Although specific tissue types, sampling strategies, and DNA or RNA extraction procedures may be needed based on the particular pathogen, genetic detection of wildlife diseases and quantification of the pathogen has become a fundamental component of wildlife disease diagnostics.

These PCR-based methods require knowing which pathogens are likely present, but disease etiology is not always known. Merely detecting the presence of a pathogen is not sufficient for many wildlife disease studies, particularly for understanding epidemiological questions related to disease transmission, rate of infection, dispersal, and evolution (Benton et al. 2015). The foundation for genetic comparison of pathogens is fairly simple: find mutations in conserved nucleotide sequences that can be used to determine how samples are related to each other. Closely related samples will share most of the same mutations, but as samples get more distantly related they will share fewer mutations. Progressively more sophisticated approaches have been developed that can sequence entire genomes of pathogens, allowing for the discovery of genetically variable sites among even very closely related samples.

New genetic approaches allow for all microbes in a sample to be potentially identified to discover likely pathogens. Although typically used to assess the human microbiome and microbial communities (Caporaso et al. 2011), amplification of conserved gene targets such as the 16S rRNA gene for bacteria and ITS/rRNA regions for fungi can be sequenced to determine the pathogens present in clinical samples (Liu 2011, Rampini et al. 2011). This approach uses a reference library to match against sequences from the sample. Because only a short amount of DNA must be sequenced for species-level identification of pathogens, this method is relatively inexpensive but is not suitable for novel pathogens whose sequences are not yet in the reference library database. More comprehensive analyses can now be achieved with metagenomic sequencing, or related approaches, that sequence all of the nucleic acids in a sample (Miller et al. 2013). Determining disease causation usually requires additional investigation, but when sequence reads are compared to genomic databases, pathogen identity, evolutionary history (Sahl et al. 2015), and traits such as antibiotic resistance can be identified (Köser et al. 2014). These new sequencing approaches generate massive amounts of data, so bioinformatic pipelines have been developed that bundle together various tools and programs to sort the data and identify the likely pathogens (Naccache et al. 2014, Rawat et al. 2014). Relatively high costs have prevented widespread use of these approaches for wildlife diseases, although decreasing sequencing prices should make them feasible, particularly for pathogen discovery of novel diseases that otherwise would remain unknown (Calistri and Palù 2015).

In particular, the application of genomic methods to wildlife disease research has drawn from advances in human diseases (Morand et al. 2012), which has culminated in whole genome analyses for public health investigations (Aarestrup et al. 2012). This same analytical paradigm can be applied to wildlife diseases and epidemiology (Archie et al. 2009), especially in pathogens with relatively limited genetic diversity, where whole genomes may be necessary to identify sufficient variation to address epidemiological questions. Considerable progress has been made in genomic analyses of some wildlife pathogens, particularly zoonotic bioterrorism agents like anthrax, a bacterium that commonly occurs in large ungulates (Kenefic et al. 2009); tularemia affecting rabbits, hares, and various rodents (Vogler et al. 2009); and plague that affects prairie dogs (*Cynomys* spp.; Morelli et al. 2010). Genomic data allowed epidemiological tracking of these diseases on a continental scale that was not feasible using older approaches. Prior to whole genome approaches, a range of different strategies have been successfully used for wildlife epidemiology including amplified-fragment length polymorphisms (AFLP) for avian diseases such as mycoplasmal conjunctivitis (Cherry et al. 2006) and avian cholera (Blehert et al. 2008). Another commonly used approach targeting mutational repeat regions in the pathogen genome, analysis of microsatellites also referred to as variable number tandem repeats (VNTRs), has revealed transmission patterns of plague (Girard et al. 2004) and the exchange of *Brucella abortus* bacteria between wildlife and livestock (Beja-Pereira et al. 2009). These earlier methods provided the foundations of molecular epidemiology in wildlife, but the analytical power of entire pathogen genomes will allow the study of wildlife disease to advance in new directions (Benton et al. 2015), including phylodynamic approaches (see below) that use genetic variation to understand pathogen transmission and spread.

DISEASE TRANSMISSION AND SPREAD

To predict the future trajectory of epidemics or successfully manage wildlife diseases, we need to understand factors that influence local disease transmission (temporal patterns) and spread (spatial distribution). Determining how and when pathogen transmission occurs, which animals become infected, and identifying heterogeneities in transmission can help managers design effective interventions. Understanding factors responsible for the spatial distribution of wildlife disease can help managers determine the geographic scope of a disease outbreak, identify populations at highest risk of infection, and design surveillance and control programs (Ostfeld et al. 2005). Rates of disease occurrence (presence–absence, prevalence, or incidence) may vary over time and space in complex and potentially interacting ways. In this section, we describe how genetic methods can contribute to elucidating mechanisms of disease transmission and spread within and among wildlife populations.

Molecular analysis of vector blood meals has revolutionized our understanding of disease transmission dynamics by the ability to identify the vertebrate hosts and pathogens being

transmitted (Kent 2009). Vectors can transmit a variety of pathogens that affect wildlife, livestock, and human hosts (e.g., plague, West Nile virus, epizootic hemorrhagic disease, malaria). Mitochondrial genes have been particularly useful targets for identifying blood meals in disease-transmitting vectors including mosquitoes transmitting West Nile and other arboviruses (Molaei et al. 2006, Hamer et al. 2009, Crabtree et al. 2013), blackflies transmitting blood parasites (Hellgren et al. 2008, 2009), and several tick-borne diseases (Gariepy et al. 2012). Ribosomal RNA markers have also been used to identify blood meals in ticks and microsatellites have been used to identify blood meals in bird-feeding mosquitoes (Darbro et al. 2007). However, there are several remaining challenges to using genetic methods for vector blood meal identification. Engorged vectors must be obtained, and undigested blood must be recovered for DNA amplification. Genetic methods for blood meal identification may also be limited by the number of potential hosts for which reference genetic data are available (Gariepy et al. 2012). Additionally, blood meals from multiple vertebrate species can pose additional difficulties.

Despite these challenges, blood meal analyses have been used to identify vector feeding preferences and successfully reconstruct likely disease transmission networks. For instance, mosquito blood meal analysis combined with knowledge of host competence was used to identify which bird species were most important in the amplification of West Nile virus in suburban Chicago, Illinois, USA (Hamer et al. 2009). Similarly, identification of blood meals in 5 bird-feeding species of black flies demonstrated strong host preferences among the flies, which may limit transmission of blood parasites among host species (Hellgren et al. 2008).

In non-vector borne diseases, contact rates among hosts can determine transmission dynamics (McCallum et al. 2001, Cross et al. 2009*b*, Craft and Caillaud 2011). For directly transmitted (animal-to-animal) diseases, constructing host contact networks for disease transmission can help managers target control efforts on particular individuals, groups, or species at higher risk of infection or disease transmission to others (White et al. 2016). Spatial aggregation of related individuals, typically exhibiting high contact rates, is a common form of social organization in wildlife (Altizer et al. 2003). Genetic markers can provide a useful tool to elucidate the influence of host relatedness on disease transmission. For instance, white-tailed deer (*Odocoileus virginianus*) with bovine tuberculosis (bTB) were more closely related than non-infected deer, suggesting that contact within family groups was a significant route of disease transmission (Blanchong et al. 2007). Similar findings have also been reported for chronic wasting disease (CWD) and relatedness in mule (*O. hemionus*) and white-tailed deer (Grear et al. 2010; Cullingham et al. 2011*a*,*b*; Magle et al. 2013). The importance of host relatedness (or contact) on disease risk, however, will depend on the type of contact, the pathogen of interest, its mechanism of transmission, social behavior of the host, and other factors (Cross et al. 2009*a*). In a study of raccoons (*Procyon lotor*) across many spatially discrete patches, the probability of 2 individuals in a patch being infected with canine distemper, a directly transmitted virus, was positively related to raccoon kin-structure (Dharmarajan et al. 2012). In contrast, familial relationships were not associated with increased disease risk for leptospirosis, caused by an environmentally transmitted bacterium. Rather, risk of leptospirosis in a patch was positively related to raccoon gene flow among patches.

In some situations, constructing temporal or spatial genetic relationships of the pathogen can help identify networks that facilitate disease transmission. Bull et al. (2012) reported that sleepy lizards (*Tiliqua rugosa*) sharing the same *Salmonella enterica* genotypes were strongly connected in the lizard social network, whereas there was no relationship between spatial proximity of hosts and bacterial genotypes. The authors concluded that *S. enterica* transmission was the result of host contacts rather than exposure to an environmental source. Building on this approach, genetic subtypes of a non-pathogenic *Escherichia coli* were used as surrogates to construct a transmission network across 10 species of wild and domestic ungulates in Kenya (VanderWaal et al. 2014), and reported Grant's gazelle (*Gazella granti*) and zebra (*Equus burchelli*) were central to multi-species transmission in this ecosystem, though in different ways. Gazelles had a large number of connections in the transmission network, suggesting they function as super-spreaders, whereas zebras seemed to play a key role in connecting different components of the network. Using microorganisms to construct transmission networks shows great promise, but it is important to remember that such an approach does not account for potential impacts of a pathogenic organism on host contact structure (Craft 2015). The amount of pathogen variation is an important consideration in identifying transmission mechanisms and contact networks. Too little genetic variation in the pathogen may lead to all individuals being equally connected, whereas too much variation will result in difficulty creating a network because most individuals possess genetically distinct pathogens. Whole genome sequence data for pathogens increasingly provide much needed depth of information as demonstrated by the elucidation of fine-scale transmission of bTB from badgers (*Meles meles*) to cattle (Biek et al. 2012) and continent-scale movement of chytrid fungus in amphibians (Rosenblum et al. 2013).

The spatial genetic structure of wildlife populations can provide insights into the spatial distribution and potential spread of disease (Kelly et al. 2010, Cullingham et al. 2011*b*, Rogers et al. 2011, Talbot et al. 2013). The spatial scale of genetic correlation among individual hosts or demographic groups can reflect the amount of gene flow that occurs (Cullingham et al. 2008). Substantial spatial structure may indicate genetic isolation among subpopulations, which suggests a reduced risk of disease spread among these populations. In contrast, a lack of spatial structure among hosts suggests higher rates of gene flow and contact among individuals across space. For example, minimal genetic structure was found among subpopulations of white-tailed deer in western Canada, highlighting the potential for long-distance spread of CWD (Cullingham et al. 2011*a*).

However, significant genetic structure occurred at fine spatial scales, suggesting that local disease transmission might occur within social groups. Vander Wal et al. (2013) examined spatial genetic structure in sympatric white-tailed deer and elk (*Cervus canadensis*) to assess the relative role of each species in the spread of bTB among elk subpopulations in Canada. The weak genetic structure in deer indicated high rates of movement among subpopulations making them likely to serve as vectors of bTB spread among strongly structured elk subpopulations.

Sex differences in genetic structure of wildlife hosts can also provide insight into mechanisms of local persistence and spatial spread of disease. Different spatial genetic patterns between nuclear genetic (maternal and paternal) markers and mitochondrial (maternal) markers can suggest differential contact and dispersal between males and females (Driscoll et al. 2015). For example, several studies in deer have documented sex-biased spatial autocorrelation in relatedness indicative of female philopatry and male dispersal. These results suggest that females primarily influence local transmission, whereas the spatial spread of disease is primarily facilitated by males (Cullingham et al. 2011*a*, Lang and Blanchong 2012). It is important to keep in mind that genetic structure in host populations is the result of host movement and interbreeding (i.e., gene flow) in the new population. Therefore, it is possible for directly transmitted diseases to spread among genetically differentiated populations if host movements occur that do not result in gene flow. For instance, in the urban landscape of southern California, Lee et al. (2012) reported that bobcat (*Lynx rufus*) subpopulations were genetically distinct, but that feline immunodeficiency virus (FIV) isolates from bobcats showed no such differentiation among subpopulations. These results are consistent with movement of bobcats among subpopulations that facilitated disease spread from feces despite minimal gene flow. Another important caveat is that gene flow among populations may reflect historical host population dynamics and may not accurately reflect contemporary or future dynamics (Epps et al. 2007). Techniques such as assignment tests and Bayesian clustering analysis applied to genetic data can directly identify migrants to gain insight into the magnitude and patterns of host movement in contemporary populations (Manel et al. 2005, Remais et al. 2011).

Population genetic approaches can also be used to track the geographic distribution and dispersal of wildlife pathogens. Tools for understanding pathogen spatial patterns include population structuring as might also be used for host populations, evolutionary (phylogenetic) trees, and geographic patterns of genetic diversity (phylogeography; Archie et al. 2009, Benton et al. 2015). A phylogenetic analysis of *Batrachochytrium dendrobatidis*, the fungus responsible for chytridiomycosis, for instance, identified a single pathogen lineage consistent with a unique geographic origin followed by a rapid spread (James et al. 2009), perhaps due to commercial movement of frogs (Fisher et al. 2009). Similarly, sequencing of the hemagglutinin gene of canine distemper virus (CDV) isolates from 16 countries over a 37-year period showed that CDV emerged in the United States in the late 1800s, subsequently diversified, and spread worldwide (Panzera et al. 2015). In addition to detecting relationships among pathogen isolates, new phylodynamic approaches using phylogenetic trees and temporal sampling have been used to infer disease transmission and spread. For instance, Kamath et al. (2016) used 245 genomes of *Brucella abortus* to describe the evolution, transmission, and timing of spatial spread of brucellosis among livestock, bison (*Bison bison*), and elk in the Greater Yellowstone ecosystem, uncovering multiple introductions of the disease, asymmetric transmission among species, and a more recent emergence of the pathogen than predicted by historical records. Phylogeographic techniques have been important in characterizing avian influenza (AI) virus diversity, gene flow among wild birds, and inferring the importance of geographic distance and avian flyway on the spread of AI across North America (Lam et al. 2012). Molecular characterization of AI viruses in mallards (*Anas platyrhynchos*) wintering in California indicated migrant birds carried variants from the Pacific Rim and Alaska, facilitating continental gene flow, whereas resident birds were responsible for perpetuating year-round transmission of several of these genetic variants (Hill et al. 2012).

Because of their relatively rapid rate of evolution, viruses can reveal patterns of host movement and provide insight into patterns of disease distribution and spread that may not be apparent in the host genetic data. Sequence information from isolates of FIV from mountain lions (*Puma concolor*) across the Rocky Mountain region of the United States and Canada exhibited pronounced spatial genetic structure providing information on the recent demographic history of mountain lions that was not evident in the mountain lion microsatellite data (Biek et al. 2006). Characterizing phylogeographic patterns in pathogens can also reveal how host population dynamics shape pathogen evolution. Some populations of bobcats in North America are infected with clade A FIV (FIVA), whereas mountain lions are primarily infected by clade B virus (FIVB; Franklin et al. 2007, Pecon-Slattery et al. 2008). The phylogeography of FIVA exhibits strong spatial structure consistent with the relatively short dispersal distances and genetic structuring of its primary host, the bobcat (Lee et al. 2014). In contrast, the phylogeography of FIVB isolates exhibit complex genetic structure; genetically diverse isolates co-circulate in some areas, whereas some genetically related isolates are found thousands of kilometers apart (Lee et al. 2014). These findings are consistent with the high mobility of FIVB's mountain lion host.

Integrating population genetics with landscape features or habitat composition, called landscape genetics (Manel et al. 2003), can further enhance our ability to understand and identify factors that influence wildlife dispersal and disease distribution. An improved understanding of these processes can be an important tool in predicting the distance and direction of disease spread (Biek and Real 2010). Management programs to contain or eradicate disease may be more effective if they incorporate or enhance natural barriers to

disease spread (Russell et al. 2006, Wheeler et al. 2010). The spatial distribution of rabies illustrates the effects of landscape features on disease distribution. Raccoon rabies prevalence in eastern Ontario, for example, was highly correlated with the degree to which different rivers acted as barriers to raccoon gene flow (Cullingham et al. 2009). Landscape features including rivers, highways, and mountain ranges have also been associated with reduced gene flow in deer and the distribution of CWD (Blanchong et al. 2008, Cullingham et al. 2011*a*, Lang and Blanchong 2012, Robinson et al. 2012*b*, Kelly et al. 2014). Incorporating landscape genetics and landscape ecology into epidemiological models may further enhance our ability to explain the current spatial distribution of disease and result in improved prediction of future spread. Robinson et al. (2013) reported that including landscape barriers to deer dispersal, identified through landscape genetics, significantly improved their predictive power to model the spatial distribution of CWD in southern Wisconsin and northern Illinois. However, in other situations, landscape features may not limit animal movement, and thus disease spread. DeYoung et al. (2009), for example, were unable to identify natural boundaries to gene flow, and thus rabies spread, for gray fox (*Urocyon cinereoargenteus*) in Texas. Their results indicate oral rabies vaccination needs to be spatially extensive to contain rabies spread in foxes.

HOST SUSCEPTIBILITY AND PATHOGEN VIRULENCE

Virulence of pathogens and susceptibility of wildlife species to disease are influenced by attributes of the host and the pathogen. Discovering the factors that determine host susceptibility to disease and how these interact with pathogen virulence are critical to understanding transmission, evaluating evolutionary consequences, and developing strategies to mitigate the effect of disease on wildlife populations. In this section, we describe how factors affecting host resistance (i.e., the ability to reduce or eliminate the probability of pathogen infection) or tolerance (i.e., the ability to limit disease severity for a given pathogen burden) to disease can be elucidated using genetic methods. Together, resistance and tolerance are the main components of host defense that determine disease severity (Raberg et al. 2007). Resistance and tolerance can also have different implications for the pathogen. For instance, resistance tends to negatively affect pathogen abundance, whereas tolerance does not, leading to potentially different evolutionary outcomes in host–pathogen interactions (Boots et al. 2009). In addition to host susceptibility, pathogen frequency and genetic diversity also can significantly influence pathogen virulence and affect host fitness.

Previous studies have reported that genetic diversity of individuals and populations of wildlife, at both neutral loci and functional genes, are related to disease tolerance or resistance. Populations with high levels of inbreeding and low levels of neutral genetic diversity have reduced adaptive potential and may be more susceptible to disease (Spielman et al. 2004, Whiteman et al. 2006). In several cases, reduced levels of heterozygosity at neutral microsatellite markers have been linked to increased pathogen susceptibility in wildlife (Coltman et al. 1999, Cassinello et al. 2001, Acevedo-Whitehouse et al. 2003). Higher genetic heterozygosity (a measure of genetic diversity) was associated with decreased probability of infection and slower disease progression of bTB in wild boar (*Sus scrofa*) in Spain (Acevedo-Whitehouse et al. 2005). One explanation for this association may be that genetically diverse individuals have superior immune competence (Orme 2004). Alternatively, these neutral loci may be linked to loci under selection (Hansson and Westerberg 2002). Many studies, however, fail to find associations between neutral genetic diversity and disease susceptibility (Schwensow et al. 2007, Worley et al. 2010, Talbot et al. 2013, Osborne et al. 2015), perhaps because diversity at neutral markers cannot provide direct information on selective processes affecting host–pathogen interactions.

The major histocompatibility complex (MHC), a highly polymorphic family of vertebrate genes involved in initiation and regulation of the immune response, has been the target of considerable investigation of disease resistance and tolerance in wildlife (Bernatchez and Landry 2003, Acevedo-Whitehouse and Cunningham 2006). Several studies have reported relationships between MHC diversity and disease (Hedrick et al. 2001, Froeschke and Sommer 2005, Oliver et al. 2009, Niskanen et al. 2014). Transmission of facial tumors among Tasmanian devils, for example, is linked to reduced diversity at MHC loci because the tumor is apparently not recognized as foreign tissue by the devil's immune system (Siddle et al. 2007). In other cases, individual MHC alleles may be more important for resistance or tolerance to a specific disease than overall diversity of MHC genes (Schwensow et al. 2007, Fernandez-de-Mera et al. 2009, Savage and Zamudio 2011, Sepil et al. 2013, Niskanen et al. 2014). Westerdahl et al. (2012), for instance, found great reed warblers (*Acrocephalus arundinaceus*) with a specific MHC allele had lower malaria parasite loads. In some cases, the same MHC allele may be related to resistance to one pathogen and susceptibility to another, as was found for different blood parasites (*Plasmodium* vs. *Haemoproteus*) in house sparrows (*Passer domesticus*; Loiseau et al. 2008). There may also be variation among populations in which MHC alleles are associated with disease. For instance, Loiseau et al. (2011) reported that the MHC allele associated with avian malaria varied across house sparrow populations. It is important to note that variation at MHC loci is often extremely high; therefore, the level of MHC diversity relative to sample size can make it challenging to identify effects associated with a single genetic variant. Researchers are often forced to cluster alleles to have sufficient statistical power for analysis.

In addition to studies on MHC genes, other candidate genes have been investigated for associations with disease or immune competence in wildlife. For instance, Turner et al. (2012) reported evidence that pathogen-influenced selection maintains genetic diversity in cytokines, genes critical for initiating and mediating the immune response, in a

population of field voles (*Microtus agrestis*). Three single nucleotide polymorphisms (SNPs) located in genes with predicted immune function were associated with bTB in African buffalo (*Syncerus caffer*; le Roex et al. 2013). Using a different genetic approach, Browning et al. (2014) reported a microsatellite locus in a heparanase gene, implicated in multiple human cancers, was associated with carcinoma in sea lions (*Phocarctos hookeri*). Capitalizing on the development of genomic resources for other carnivore species, McCarthy et al. (2011) explored the association between phocine distemper in harbor seals (*Phoca vitulina*) and 8 candidate genes that encoded proteins involved with host cellular interactions with *Morbilliviruses*. Many studies have examined the relationship between amino acid polymorphisms in the prion protein (PRNP) gene and CWD infection in cervids (O'Rourke et al. 1999, Kelly et al. 2008, Perucchini et al. 2008, Blanchong et al. 2009, Wilson et al. 2009). Amino acid variation in the PRNP gene has been linked to the probability of infection with CWD (resistance) and the progression of disease following infection (tolerance; Fox et al. 2006; Johnson et al. 2006, 2011; Robinson et al. 2012*a*). These genetic associations between pathogens and host susceptibility have significant consequences for host population demography and evolution (Robinson et al. 2012*a*).

Because of the limited resources for genomic analyses for most wildlife species and wildlife diseases, genetic investigations of disease resistance or tolerance have primarily focused on a priori candidate genes suspected to be important in the disease process. The availability of genomic information for a closely related domestic counterpart can facilitate genome-wide approaches to identify genes associated with wildlife disease. For instance, Matsumoto et al. (2013) used association mapping with 215 microsatellites from a high-density genetic map of the cow genome to identify potential CWD risk factors in mule and white-tailed deer. As genomic technologies continue to advance and costs become more affordable for wildlife projects, we expect to see more genome-wide approaches, although the challenges of handling and analyzing large volumes of data produced with these approaches still remain (see Future Directions section).

Regardless of the molecular approach taken, several challenges exist in conducting observational disease association studies in free-ranging wildlife to assess disease resistance or tolerance, especially from cross-sectional data (rather than longitudinal data where individuals are followed over time) that have not been collected under an experimental framework. First, there is a lack of certainty whether all animals have the same risk of pathogen exposure. In addition, there is uncertainty whether disease-negative animals have been exposed to and cleared the pathogen versus never having been exposed. One potential improvement is to use case–control methods that match disease-negative and -positive animals based on similar disease risk factors (Blanchong et al. 2009, le Roex et al. 2013).

Pathogen diversity, virulence, and cross immunity also factor into host susceptibility and disease severity (Sorci 2013). Pathogens can have high evolutionary potential due to their large population size, high rates of replication, high genetic variation, potential for genetic reassortment, and short generation times. Pathogens typically face the significant challenges of persisting in the face of the host defense mechanisms (e.g., immune response) and successful transmission between hosts. These selection pressures frequently promote pathogen evolution. For example, the genetic variation of pathogens may be influenced by the route and rate of transmission and the host's response to infection. Waterfowl are the natural host for avian influenza (AI) viruses. The AI infection typically has few consequences for host fitness and the virus is rapidly passed among many different waterfowl species by direct and environmental routes (Henaux et al. 2013). A multi-year study of avian influenza viruses isolated from wild mallards found that individuals were rarely re-infected with the same or related hemagglutinin (HA) AI subtypes, suggesting cross-protective immunity (Latorre-Margalef et al. 2013). The need to escape the immune system appears to have selected for genetic variation (subtypes) of the AI virus. Temporal patterns in the frequency of different HA AI subtypes combined with their phylogenetic relatedness suggests herd immunity in the hosts affects virus dynamics (Latorre-Margalef et al. 2014). Echaubard et al. (2014) investigated the host–pathogen interactions of ranavirus strains of varying virulence in 2 species of amphibians. They reported that the outcome of ranavirus infection depended on the particular combination of host and viral genotype. Identification of genetic variants associated with the propensity for increased virulence or adaptation to new hosts can be an important component of disease surveillance activities. For instance, experimental studies of H5N1 and H7N7 AI viruses identified genetic mutations associated with adaptation to mammals or increased virulence (Hatta et al. 2001, Czudai-Matwich et al. 2014, Chen et al. 2015). Monitoring for changes in these amino acids during AI virus surveillance might help to identify potential situations of elevated zoonotic risk. Differential effects of various pathogen subtypes or other closely related groups highlight the need for molecular tools to identify genetic variation within pathogen genomes. Fortunately, a variety of approaches have been developed that are rapid and cost effective (see Pathogen Detection and Characterization section).

IMPACTS OF DISEASE TO HOST POPULATIONS

The coevolution of hosts and pathogens is a ubiquitous phenomenon of fundamental importance to all living organisms including wildlife. An area of current interest is whether wildlife can evolve resistance or tolerance to disease over time scales relevant to management (Boots et al. 2009). Because many wildlife pathogens are multi-host diseases, the outcome of host–pathogen coevolution can have significant implications for other affected host populations, including other wildlife species, domestic animals, and even humans. Numerous studies have reported that diseases can have significant impacts on wildlife populations, including, but not limited to, population reduction, changing age structure,

altering life-history parameters, affecting genetic diversity, and modifying behaviors (Hurtado 2008, Jones et al. 2008*b*, Lachish et al. 2009, Lee et al. 2010, Robinson et al. 2010). Although disease is one of many ecological processes that can directly influence wildlife populations, it may also predispose affected individuals to other mortality risks or otherwise indirectly reduce individual fitness (Blaustein et al. 2012). These interacting processes make it challenging to determine the population-level effects of pathogens on wildlife abundance, distribution, and demographic rates. In this section, we will review how genetic approaches can shed light on the impacts of disease to wildlife populations.

In white-tailed deer, differences in infection rates, disease progression, and survival between CWD-resistant and susceptible genotypes can produce a fitness advantage for individuals with the CWD-resistant genotype, which is predicted to increase the frequency of resistant genotype (Robinson et al. 2012*a*). The rate of change in resistant or susceptible genotypes, however, is a function of the magnitude of the fitness advantage, selection pressure, and population mixing. Based on CWD infection rates in Wisconsin deer, the CWD-resistant type is predicted to increase but not at a pace relevant for contemporary deer management. Tschirren (2015) reported similar results when examining the effects of the bacterium responsible for Lyme disease (*Borrelia burgdorferi*) in bank voles (*Myodes glareolus*). Specifically, the frequency of the protective Toll-like receptor 2 (TLR2) variant was higher in vole populations with greater infection pressure (as measured by human incidence of disease), providing evidence that *Borrelia*-mediated selection affected the frequency of this receptor. The low frequency of these protective genetic types in wildlife populations where disease is rare or recently introduced suggests that uncommon genotypes may be associated with fitness costs in the absence of disease. For example, in humans, a Toll-like receptor 4 variant is associated with reduced malaria mortality but is disadvantageous to individuals when malaria is absent because it is associated with increased susceptibility to other infections (Ferwerda et al. 2007). Evaluating the fitness costs of genetic variants associated with disease resistance or tolerance in the absence of disease in wildlife populations is an important area for future research.

Genetic methods can identify the influence of genetic diversity and disease selection pressure on the development of disease tolerance or resistance. Since its introduction, avian malaria has devastated endemic honeycreeper species in Hawaii (van Riper III et al. 1986). One species, the Hawaii amakihi (*Chlorodrepanis virens*), has shown a remarkable population recovery in lowland forests on the island of Hawaii despite the high prevalence of malaria (Woodworth et al. 2005, Samuel et al. 2015), which has prevented 2 other native species, the apapane (*Himatione sanguinea*) and iiwi (*Vestiaria coccinea*), from recolonizing the same lowland areas. Foster et al. (2007) reported that lowland amakihi are genetically distinct from other amakihi populations and hypothesized that high prevalence of disease in lowland areas rapidly selected for malaria tolerance in these birds. Subsequent laboratory and field investigations confirmed this amakihi population has much lower malaria mortality than found in apapane, iiwi, or even amakihi in other populations (Atkinson et al. 2013, Samuel et al. 2015). However, the specific gene(s) that allows this population of amakihi to tolerate malaria infection have not yet been identified. In contrast, apapane and iiwi exhibit little genetic differentiation among populations at different elevations. The high rate of movement (gene flow) of apapane and iiwi among elevations, combined with a strong elevational gradient in malaria infection pressure (Samuel et al. 2015), may have limited the development of disease tolerance in these species, preventing them from recolonizing areas where avian malaria infection is high.

Genetic methods have also been used to understand potential impacts of disease on wildlife dispersal and related population processes that may be difficult to elucidate with other techniques. Several studies have investigated the effects of disease on genetic diversity and structure in wildlife populations that may be the result of altered dispersal patterns or other behavioral changes (Trudeau et al. 2004, Teacher et al. 2009). For instance, changes in population density associated with Tasmanian devil facial tumor disease appear to have reshaped dispersal patterns that are now reflected in altered gene flow among populations (Lachish et al. 2011, Bruniche-Olsen et al. 2013). Serieys et al. (2015) used neutral genetic markers to show that an epizootic of mange in California bobcats resulted in genetic differentiation between populations before and after the epizootic that was greater than between populations separated by a highway for over 60 years. The post-epidemic population had a 10-fold higher inbreeding coefficient at neutral genetic markers but a decreased inbreeding coefficient at immune-linked loci after the epidemic, suggesting disease pressure acted to maintain variation at immune function genes associated with disease.

Management actions aimed at controlling disease in wildlife populations can sometimes have unintended genetic consequences. For instance, a selective culling program to reduce bTB in African buffalo resulted in a higher frequency of rare alleles at the IFNG locus, which codes for interferon gamma, of crucial importance for immune response, compared to non-culled populations (Lane-deGraaf et al. 2015). This suggests that culling was leading to a loss of immunogenetic diversity, with potentially important evolutionary and future population consequences. Blanchong et al. (2012) used genetics to examine indirect effects of CWD on reproduction and fawn harvest vulnerability in Wisconsin deer populations. Specifically, the authors reconstructed parent–offspring relationships and found that male fawns from CWD-infected mothers may be more vulnerable to harvest than fawns from non-infected mothers. Genetics can, therefore, be used to inform the efficacy of management actions aimed to control disease.

GENOMICS—PROGRESS TO DATE AND FUTURE DIRECTIONS

Genomic methods have been used for some time to investigate the pathogen side of host–pathogen dynamics.

As a result, the genomes of numerous pathogens have been sequenced and this sequencing will become increasingly common. Additionally, development of genomic methodologies that enable detection of unknown pathogens will facilitate pathogen discovery, early detection of disease risk, and contribute to our understanding of disease emergence and spread (Lipkin 2013). Metagenomics is enabling sequencing and identification of microbial genomes present in a whole suite of sample types, including environmental and clinical samples (Doolittle and Zhaxybayeva 2010, Drexler et al. 2012). This will facilitate environmental screening for pathogens of concern prior to wildlife translocation or reintroductions, evaluation of success following management actions to control pathogens, or identification of potential zoonotic risks. For instance, Baker et al. (2013) used metagenomics to identify several novel viruses in urine from bats roosting in close proximity to humans. Microbiome studies (Grice and Segre 2011) are being conducted on bats (Johnson et al. 2013) to identify host-specific microorganisms and their potential role in facilitating or inhibiting disease transmission (Hooper et al. 2012). In the future, field-based sequencing of pathogens might enable managers to conduct more rapid and effective pathogen surveillance, evaluate disease risk, and monitor pathogen exposure in wildlife populations. For pathogens that evolve rapidly, methods such as phylodynamics (Grenfell et al. 2004), which combines phylogenetics, epidemiology, population genetics, and immunology (Volz et al. 2013), can be used to assess their potential evolutionary and epidemiological dynamics. As a result of these novel and interdisciplinary genetic approaches, our understanding of transmission and temporal and spatial spread of wildlife diseases will be enhanced.

In contrast to pathogen genomics, many studies on disease dynamics in wildlife populations have relied on a limited number of neutral genetic markers. Neutral markers are an indirect means of investigating relationships between host genetics and disease susceptibility. For instance, the use of 10–30 microsatellite markers captures a small fraction of a host genome, and thus does not necessarily represent genome-wide genetic diversity (Vali et al. 2008). This may partially explain differing results among studies investigating associations between neutral genetic diversity and disease susceptibility. Moreover, although neutral markers provide an excellent understanding of how gene flow and genetic drift shape population genetic structure, they do not provide insight on fitness or genetic adaptation across the landscape, and therefore cannot identify how selective forces, such as pathogens, drive adaptation (Manel and Holderegger 2013). The increasing availability and affordability of next-generation sequencing will facilitate genome-wide association studies (GWAS) that can identify host genes or genome regions associated with resistance and tolerance to disease (Segura et al. 2012). For example, the recently sequenced genome of the amakihi, the only Hawaiian honeycreeper with known tolerance to avian malaria, identified roughly 3.9 million SNPs (Callicrate et al. 2014), which provides a platform to discover SNPs associated with disease in this species as well as other species of honeycreepers threatened by malaria. Genomic approaches are also increasingly available for wildlife species for which no genomic sequence information is available (or do not have a closely related species whose genome has been sequenced). For example, approaches that rely on restriction enzymes (Davey et al. 2011) such as restriction-site-associated DNA sequencing (RAD-seq) or genotyping by sequencing (GBS) can be applied to any organism and may aid in efforts to identify genetic variants associated with disease status. Genomic data will enable epidemiological studies to identify variants at candidate immune or other genes hypothesized to be directly involved in resistance or tolerance (Brown and Knowles 2012). These findings will contribute to our ability to predict the outcome of host–pathogen interactions, develop prevention strategies, and evaluate the effects of management actions on evolutionary processes.

The emerging discipline of landscape epidemiology, which combines the fields of landscape ecology and landscape genetics, recognizes that landscape complexity plays a significant role in the spatial distribution and spread of diseases. In general, the goal of landscape epidemiology is to understand and predict patterns of disease occurrence and risk across complex landscapes (Ostfeld et al. 2005). Further, the expansion of landscape genetics to landscape genomics may help researchers determine how genetic variation across the landscape is directly related to disease risk, susceptibility, or immune competence, and assess environmental heterogeneity related to selection acting on both hosts and pathogens. Innovative approaches that involve integrating host and pathogen phylogenies with landscape features can make novel contributions to understanding how host demographics, host genetics, and landscape features affect pathogen transmission and spread. The ability to track wildlife using increasingly smaller, longer lasting, and more complex instrumentation (e.g., transmitters) is also being integrated into emerging genomic technologies. Wildlife are being fitted with transmitters that allow tracking of contact rates, heart rates, temperature, light, and video, which combined with pathogen and host genomics can be used to understand animal movement and disease transmission across the landscape (Shafer et al. 2016).

Another emerging area in wildlife epidemiology is the use of gene expression studies to facilitate a deeper understanding of host–pathogen interactions (Westermann et al. 2012) and contribute to the field of wild immunology, which aims to apply immunology to natural populations to better understand immune mechanisms influencing host health and fitness (Pedersen and Babayan 2011). Patterns of gene expression are focused on alterations in gene activities that induce changes in biological processes. Because most diseases are thought to involve complex interactions between many genetic loci, gene expression can help determine how animals respond to pathogens. Transcriptome data, gene expression measured genome wide, can provide insight into how gene function in hosts and pathogens mediates disease dynamics.

Among other things, transcriptome data may elucidate complexities of the host immune response to infection, provide insight into the means by which pathogens evade the immune system, and identify appropriate targets for vaccines. For example, comparison of profiles between bTB-infected and non-infected European wild boar identified differences in gene expression associated with cellular processes involved with the immune response (Galindo et al. 2009), shedding light on how the bTB bacterium evades the immune system.

Although the rapid advances in technological capabilities make next-generation sequencing approaches increasingly feasible and cost-effective for wildlife disease investigations, significant challenges remain (Luikart et al. 2003, Steiner et al. 2013, Shafer et al. 2015). Data management and analysis are important challenges associated with the vastly larger datasets produced from genomic approaches. Whole genome sequencing produces enormous amounts of data that are computationally difficult and expensive to analyze and can be costly to store. This is especially true for the vertebrate hosts whose genomes are often orders of magnitude larger than the genomes of their pathogens. Cloud-based data management and storage solutions may aid in data handling (Baker 2010).

MANAGEMENT IMPLICATIONS

Wildlife epidemiology requires a multidisciplinary approach to understand the complexity of disease in wildlife. Understanding the causes of disease, transmission rates and routes, pathophysiology, susceptibility, virulence, and the ecology of pathogens and how they interact with wildlife hosts is essential for developing effective strategies to prevent or manage disease. Better understanding of these concepts will enable wildlife managers and scientists to address future disease challenges. For wildlife diseases, this approach will involve a synthesis of classical and cutting edge technologies from diverse disciplines including wildlife ecology, conservation biology, environmental biology, high-tech animal instrumentation, veterinary medicine, pathology, microbiology, and molecular biology. With this increasing complexity and interdisciplinary work, strong and broad collaborations among those with expertise in the disciplines listed above are key. For example, collaborations between disease ecologists and bioinformatics experts will be critical for the successful analysis and appropriate interpretation of the enormous amounts of data generated in next-generation sequencing studies of wildlife epidemiology. Genetics and genomics can contribute by providing tools to detect and characterize pathogens, uncover routes of disease transmission and spread, shed light on the ways that disease susceptibility is influenced by both host and pathogen attributes, and elucidate the impacts of disease on wildlife populations. Greater understanding of the complex interactions between pathogens and free-ranging wildlife will aid managers' efforts to identify future disease risks, prevent disease introduction, and mitigate the impacts of wildlife diseases for the benefit of global health and the conservation of biodiversity.

ACKNOWLEDGMENTS

We thank E. H. Merrill for stimulating this review. H. B, Ernest, R. B. Gagne, N. M. Fountain-Jones, and 2 anonymous reviewers provided many helpful comments that improved the paper. Mention of trade names or products does not imply endorsement by the U.S. Government.

LITERATURE CITED

Aarestrup, F. M., E. W. Brown, C. Detter, P. Gerner-Smidt, M. W. Gilmour, D. Harmsen, R. S. Hendriksen, R. Hewson, D. L. Heymann, K. Johansson, K. Ijaz, P. S. Keim, M. Koopmans, A. Kroneman, D. Lo Fo Wong, O. Lund, D. Palm, P. Sawanpanyalert, J. Sobel, and J. Schlundt. 2012. Integrating genome-based informatics to modernize global disease monitoring, information sharing, and response. Emerging Infectious Diseases 18:e1.

Acevedo-Whitehouse, K., and A. A. Cunningham. 2006. Is MHC enough for understanding wildlife immunogenetics? Trends in Ecology & Evolution 21:433–438.

Acevedo-Whitehouse, K., F. Gulland, D. Greig, and W. Amos. 2003. Disease susceptibility in California sea lions. Nature 422:35.

Acevedo-Whitehouse, K., J. Vicente, C. Gortazar, U. Hofle, I. G. Fernandez-De-Mera, and W. Amos. 2005. Genetic resistance to bovine tuberculosis in the Iberian wild boar. Molecular Ecology 14:3209–3217.

Altizer, S., C. L. Nunn, P. H. Thrall, J. L. Gittleman, J. Antonovics, A. A. Cunningham, A. P. Dobson, V. Ezenwa, K. E. Jones, A. B. Pedersen, M. Poss, and J. R. C. Pulliam. 2003. Social organization and parasite risk in mammals: integrating theory and empirical studies. Annual Review of Ecology, Evolution, and Systematics 34:517–547.

Archie, E. A., G. Luikart, and V. O. Ezenwa. 2009. Infecting epidemiology with genetics: a new frontier in disease ecology. Trends in Ecology & Evolution 24:21–30.

Atkinson, C. T., K. S. Saili, R. B. Utzurrum, and S. I. Jarvi. 2013. Experimental evidence for evolved tolerance to avian malaria in a wild population of low wlevation Hawai'i 'Amakihi (*Hemignathus virens*). EcoHealth 10:366–375.

Baker, K. S., R. M. Leggett, N. H. Bexfield, M. Alston, G. Daly, S. Todd, M. Tachedjian, C. E. G. Holmes, S. Crameri, L. F. Wang, J. L. Heeney, R. Suu-Ire, P. Kellam, A. A. Cunningham, J. L. N. Wood, M. Caccamo, and P. R. Murcia. 2013. Metagenomic study of the viruses of African straw-coloured fruit bats: detection of a chiropteran poxvirus and isolation of a novel adenovirus. Virology 441:95–106.

Baker, M. 2010. Next-generation sequencing: adjusting to data overload. Nature Methods 7:495–499.

Beja-Pereira, A., B. Bricker, S. Chen, C. Almendra, P. J. White, and G. Luikart. 2009. DNA genotyping suggests that recent brucellosis outbreaks in the Greater Yellowstone Area originated from elk. Journal of Wildlife Diseases 45:1174–1177.

Benton, C. H., R. J. Delahay, H. Trewby, and D. J. Hodgson. 2015. What has molecular epidemiology ever done for wildlife disease research? Past contributions and future directions. European Journal of Wildlife Research 61:1–16.

Bernatchez, L., and C. Landry. 2003. MHC studies in nonmodel vertebrates: what have we learned about natural selection in 15 years? Journal of Evolutionary Biology 16:363–377.

Biek, R., A. J. Drummond, and M. Poss. 2006. A virus reveals population structure and recent demographic history of its carnivore host. Science 311:538–541.

Biek, R., A. O'Hare, D. Wright, T. Mallon, C. McCormick, R. J. Orton, S. McDowell, H. Trewby, R. A. Skuce, and R. R. Kao. 2012. Whole genome sequencing reveals local transmission patterns of *Mycobacterium bovis* in sympatric cattle and badger populations. PLoS Pathogens 8:e1003008.

Biek, R., and L. A. Real. 2010. The landscape genetics of infectious disease emergence and spread. Molecular Ecology 19:3515–3531.

Black, E. M., J. P. Lowings, J. Smith, P. R. Heaton, and L. M. McElhinney. 2002. A rapid RT-PCR method to differentiate six established genotypes of rabies and rabies-related viruses using TaqMan technology. Journal of Virological Methods 105:25–35.

Blanchong, J. A., D. A. Grear, B. V. Weckworth, D. P. Keane, K. T. Scribner, and M. D. Samuel. 2012. Effects of chronic wasting disease on

reproduction and fawn harvest vulnerability in Wisconsin white-tailed deer. Journal of Wildlife Diseases 48:361–370.
Blanchong, J. A., D. M. Heisey, K. T. Scribner, S. V. Libants, C. Johnson, J. M. Aiken, J. A. Langenberg, and M. D. Samuel. 2009. Genetic susceptibility to chronic wasting disease in free-ranging white-tailed deer: complement component C1q and Prnp polymorphisms. Infection Genetics and Evolution 9:1329–1335.
Blanchong, J. A., M. D. Samuel, K. T. Scribner, B. V. Weckworth, J. A. Langenberg, and K. B. Filcek. 2008. Landscape genetics and the spatial distribution of chronic wasting disease. Biology Letters 4:130–133.
Blanchong, J. A., K. T. Scribner, A. N. Kravchenko, and S. R. Winterstein. 2007. TB-infected deer are more closely related than non-infected deer. Biology Letters 3:103–105.
Blaustein, A. R., S. S. Gervasi, P. T. J. Johnson, J. T. Hoverman, L. K. Belden, P. W. Bradley, and G. Y. Xie. 2012. Ecophysiology meets conservation: understanding the role of disease in amphibian population declines. Philosophical Transactions of the Royal Society of London B: Biological Sciences 367:1688–1707.
Blehert, D. S., K. L. Jefferson, D. M. Heisey, M. D. Samuel, B. M. Berlowski, and D. J. Shadduck. 2008. Using amplified fragment length polymorphism analysis to differentiate isolates of *Pasteurella multocida* serotype 1. Journal of Wildlife Diseases 44:209–225.
Boots, M., A. Best, M. R. Miller, and A. White. 2009. The role of ecological feedbacks in the evolution of host defence: what does theory tell us? Philosophical Transactions of the Royal Society of London B: Biological Sciences 364:27–36.
Boyle, D. G., D. B. Boyle, V. Olsen, J. A. Morgan, and A. D. Hyatt. 2004. Rapid quantitative detection of chytridiomycosis (*Batrachochytrium dendrobatidis*) in amphibian samples using real-time Taqman PCR assay. Diseases of Aquatic Organisms 60:141–148.
Bricker, B. J. 2004. Molecular diagnostics of animal brucellosis: a review of PCR-based assays and approaches. Pages 24–51 *in* I. Lopez-Goni, and I. Moriyon, editors. Brucella: molecular and cellular biology. Horizon Bioscience, Norfolk, United Kingdom.
Brown, J. L., and L. L. Knowles. 2012. Spatially explicit models of dynamic histories: examination of the genetic consequences of Pleistocene glaciation and recent climate change on the American Pika. Molecular Ecology 21:3757–3775.
Browning, H. M., K. Acevedo-Whitehouse, F. M. D. Gulland, A. J. Hall, J. Finlayson, M. P. Dagleish, K. J. Billington, K. Colegrove, and J. A. Hammond. 2014. Evidence for a genetic basis of urogenital carcinoma in the wild California sea lion. Proceedings of the Royal Society B-Biological Sciences 281:20140240.
Bruniche-Olsen, A., C. P. Burridge, J. J. Austin, and M. E. Jones. 2013. Disease induced changes in gene flow patterns among Tasmanian devil populations. Biological Conservation 165:69–78.
Bull, C. M., S. S. Godfrey, and D. M. Gordon. 2012. Social networks and the spread of Salmonella in a sleepy lizard population. Molecular Ecology 21:4386–4392.
Calistri, A., and G. Palù. 2015. Unbiased next-generation sequencing and new pathogen discovery: undeniable advantages and still-existing drawbacks. Clinical Infectious Diseases 60:889–891.
Callicrate, T., R. Dikow, J. W. Thomas, J. C. Mullikin, E. D. Jarvis, R. C. Fleischer, and N. C. Sequencing. 2014. Genomic resources for the endangered Hawaiian honeycreepers. BMC Genomics 15:1098.
Caporaso, J. G., C. L. Lauber, W. A. Walters, D. Berg-Lyons, C. A. Lozupone, P. J. Turnbaugh, N. Fierer, and R. Knight. 2011. Global patterns of 16S rRNA diversity at a depth of millions of sequences per sample. Proceedings of the National Academy of Sciences of the United States of America 108 Supplement 1:4516–4522.
Cassinello, J., M. Gomendio, and E. R. S. Roldan. 2001. Relationship between coefficient of inbreeding and parasite burden in endangered gazelles. Conservation Biology 15:1171–1174.
Chen, Q., Z. J. Yu, W. Y. Sun, H. L. Chai, X. L. Gao, J. Guo, K. Zhang, N. Feng, X. X. Zheng, H. L. Wang, Y. K. Zhao, C. Qin, G. Huang, S. T. Yang, J. Qian, Y. W. Gao, X. Z. Xia, T. C. Wang, and Y. P. Hua. 2015. Adaptive amino acid substitutions enhance the virulence of H7N7 avian influenza virus isolated from wild waterfowl in mice. Veterinary Microbiology 177:18–24.
Cherry, J. J., D. H. Ley, and S. Altizer. 2006. Genotypic analyses of *Mycoplasma gallisepticum* isolates from songbirds by random amplification of polymorphic DNA and amplified-fragment length polymorphism. Journal of Wildlife Diseases 42:421–428.
Cohen, M. L. 2000. Changing patterns of infectious disease. Nature 406:762–767.
Coltman, D. W., J. G. Pilkington, J. A. Smith, and J. M. Pemberton. 1999. Parasite-mediated selection against inbred Soay sheep in a free-living, island population. Evolution 53:1259–1267.
Crabtree, M. B., R. C. Kading, J.-P. Mutebi, J. J. Lutwama, and B. R. Miller. 2013. Identification of host blood from engorged mosquitoes collected in western Uganda using cytochrome oxidase I gene sequences. Journal of Wildlife Diseases 49:611–626.
Craft, M. E. 2015. Infectious disease transmission and contact networks in wildlife and livestock. Philosophical Transactions of the Royal Society B-Biological Sciences 370:20140107.
Craft, M. E., and D. Caillaud. 2011. Network models an underutilized tool in wildlife epidemiology. Interdisciplinary Perspectives on Infectious Diseases 2011:676949.
Cross, P. C., D. M. Heisey, J. A. Bowers, C. T. Hay, J. Wolhuter, P. Buss, M. Hofmeyr, A. L. Michel, R. G. Bengis, T. L. F. Bird, J. T. Du Toit, and W. M. Getz. 2009*a*. Disease, predation and demography: assessing the impacts of bovine tuberculosis on African buffalo by monitoring at individual and population levels. Journal of Applied Ecology 46:467–475.
Cross, P. C., J. Drewe, V. Patrek, G. Pearce, M. D. Samuel, and R. J. Delahay. 2009*b*. Host population structure and implications for disease management. Pages 9–29 *in* R. J. Delahay, G. C. Smith, and M. R. Hutchings, editors. Management of disease in wild mammals. Springer-Verlag Tokyo, Inc., Tokyo, Japan.
Cullingham, C. I., C. J. Kyle, B. A. Pond, E. E. Rees, and B. N. White. 2009. Differential permeability of rivers to raccoon gene flow corresponds to rabies incidence in Ontario, Canada. Molecular Ecology 18:43–53.
Cullingham, C. I., C. J. Kyle, B. A. Pond, and B. N. White. 2008. Genetic structure of raccoons in Eastern North America based on mtDNA: implications for subspecies designation and rabies disease dynamics. Canadian Journal of Zoology-Revue Canadienne De Zoologie 86:947–958.
Cullingham, C. I., E. H. Merrill, M. J. Pybus, T. K. Bollinger, G. A. Wilson, and D. W. Coltman. 2011*a*. Broad and fine-scale genetic analysis of white-tailed deer populations: estimating the relative risk of chronic wasting disease spread. Evolutionary Applications 4:116–131.
Cullingham, C. I., S. M. Nakada, E. H. Merrill, T. K. Bollinger, M. J. Pybus, and D. W. Coltman. 2011*b*. Multiscale population genetic analysis of mule deer (*Odocoileus hemionus hemionus*) in western Canada sheds new light on the spread of chronic wasting disease. Canadian Journal of Zoology-Revue Canadienne De Zoologie 89:134–147.
Czudai-Matwich, V., A. Otte, M. Matrosovich, G. Gabriel, and H. D. Klenk. 2014. PB2 mutations D701N and S714R promote adaptation of an influenza H5N1 virus to a mammalian host. Journal of Virology 88:8735–8742.
Darbro, J. M., A. A. Dhondt, F. M. Vermeylen, and L. C. Harrington. 2007. *Mycoplasma gallisepticum* infection in house finches (*Carpodacus mexicanus*) affects mosquito blood feeding patterns. American Journal of Tropical Medicine and Hygiene 77:488–494.
Daszak, P., A. A. Cunningham, and A. D. Hyatt. 2000. Wildlife ecology—emerging infectious diseases of wildlife—threats to biodiversity and human health. Science 287:443–449.
Davey, J. W., P. A. Hohenlohe, P. D. Etter, J. Q. Boone, J. M. Catchen, and M. L. Blaxter. 2011. Genome-wide genetic marker discovery and genotyping using next-generation sequencing. Nature Reviews Genetics 12:499–510.
Decaro, N., G. Elia, V. Martella, C. Desario, M. Campolo, L. D. Trani, E. Tarsitano, M. Tempesta, and C. Buonavoglia. 2005. A real-time PCR assay for rapid detection and quantitation of canine parvovirus type 2 in the feces of dogs. Veterinary Microbiology 105:19–28.
Deem, S. L., W. B. Karesh, and W. Weisman. 2001. Putting theory into practice: wildlife health in conservation. Conservation Biology 15:1224–1233.
Delaney, N. F., S. Balenger, C. Bonneaud, C. J. Marx, G. E. Hill, N. Ferguson-Noel, P. Tsai, A. Rodrigo, and S. V. Edwards. 2012. Ultrafast evolution and loss of CRISPRs following a host shift in a novel wildlife pathogen, *Mycoplasma gallisepticum*. PLoS Genetics 8:e1002511.
DeYoung, R. W., A. Zamorano, B. T. Mesenbrink, T. A. Campbell, B. R. Leland, G. M. Moore, R. L. Honeycutt, and J. J. Root. 2009. Landscape-

genetic analysis of population structure in the Texas gray fox oral rabies vaccination zone. Journal of Wildlife Management 73:1292–1299.
Dharmarajan, G., J. C. Beasley, J. A. Fike, E. A. Raizman, C. C. Wu, R. M. Pogranichniy, and O. E. Rhodes. 2012. Effects of kin-structure on disease dynamics in raccoons (*Procyon lotor*) inhabiting a fragmented landscape. Basic and Applied Ecology 13:560–567.
Dobson, A., and J. Foufopoulos. 2001. Emerging infectious pathogens of wildlife. Philosophical Transactions of the Royal Society of London Series B-Biological Sciences 356:1001–1012.
Doolittle, W. F., and O. Zhaxybayeva. 2010. Metagenomics and the units of biological organization. Bioscience 60:102–112.
Drexler, J. F., V. M. Corman, M. A. Muller, G. D. Maganga, P. Vallo, T. Binger, F. Gloza-Rausch, V. M. Cottontail, A. Rasche, S. Yordanov, A. Seebens, M. Knornschild, S. Oppong, Y. Adu Sarkodie, C. Pongombo, A. N. Lukashev, J. Schmidt-Chanasit, A. Stocker, A. J. Carneiro, S. Erbar, A. Maisner, F. Fronhoffs, R. Buettner, E. K. Kalko, T. Kruppa, C. R. Franke, R. Kallies, E. R. Yandoko, G. Herrler, C. Reusken, A. Hassanin, D. H. Kruger, S. Matthee, R. G. Ulrich, E. M. Leroy, and C. Drosten. 2012. Bats host major mammalian paramyxoviruses. Nature Communications 3:796.
Driscoll, C. C., J. G. Driscoll, C. Hazekamp, J. B. Mitton, and J. D. Wehausen. 2015. A tale of two markers: population genetics of Colorado rocky mountain bighorn sheep estimated from microsatellite and mitochondrial data. Journal of Wildlife Management 79:819–831.
Echaubard, P., J. Leduc, B. Pauli, V. G. Chinchar, J. Robert, and D. Lesbarreres. 2014. Environmental dependency of amphibian-ranavirus genotypic interactions: evolutionary perspectives on infectious diseases. Evolutionary Applications 7:723–733.
Eldridge, B. F., and J. D. Edman, editors. 2004. Medical entomology: a textbook on public health and veterinary problems caused by arthropods. Springer, Dordrecht, Netherlands.
Epps, C. W., J. D. Wehausen, V. C. Bleich, S. G. Torres, and J. S. Brashares. 2007. Optimizing dispersal and corridor models using landscape genetics. Journal of Applied Ecology 44:714–724.
Fernandez-de-Mera, I. G., J. Vicente, V. Naranjo, Y. Fierro, J. J. Garde, J. de la Fuente, and C. Gortazar. 2009. Impact of major histocompatibility complex class II polymorphisms on Iberian red deer parasitism and life history traits. Infection Genetics and Evolution 9:1232–1239.
Ferwerda, B., M. B. B. McCall, S. Alonso, M. Mouktaroudi, E. J. Giamarellos-Bourboulis, N. Izagirre, D. Syafruddin, G. Kibiki, T. Cristea, A. Hijmans, L. Hamann, S. Israel, G. Eighazali, M. Troye-Blomberg, O. Kumpf, B. Maiga, A. Dolo, O. Doumbo, C. C. Hermsen, A. F. H. Stalenhoef, R. van Crevel, H. G. Brunner, D. Y. Oh, R. R. Schumann, C. de la Rua, R. Sauerwein, B. J. Kullberg, A. J. A. M. van der Ven, J. W. M. van der Meer, and M. G. Netea. 2007. TLR4 polymorphisms, infectious diseases, and evolutionary pressure during migration of modern humans. Proceedings of the National Academy of Sciences of the United States of America 104:16645–16650.
Fisher, M. C., T. W. Garner, and S. F. Walker. 2009. Global emergence of *Batrachochytrium dendrobatidis* and amphibian chytridiomycosis in space, time, and host. Annual Reviews in Microbiology 63:291–310.
Foster, J. T., B. L. Woodworth, L. E. Eggert, P. J. Hart, D. Palmer, D. C. Duffy, and R. C. Fleischer. 2007. Genetic structure and evolved malaria resistance in Hawaiian honeycreepers. Molecular Ecology 16:4738–4746.
Fox, K. A., J. E. Jewell, E. S. Williams, and M. W. Miller. 2006. Patterns of Prp(CWD) accumulation during the course of chronic wasting disease infection in orally inoculated mule deer (*Odocoileus hemionus*). Journal of General Virology 87:3451–3461.
Franklin, S. P., J. L. Troyer, J. A. Twrwee, L. M. Lyren, W. M. Boyce, S. P. D. Riley, M. E. Roelke, K. R. Crooks, and S. VandeWoude. 2007. Frequent transmission of immunodeficiency viruses among bobcats and pumas. Journal of Virology 81:10961–10969.
Frick, W. F., J. F. Pollock, A. C. Hicks, K. E. Langwig, D. S. Reynolds, G. G. Turner, C. M. Butchkoski, and T. H. Kunz. 2010. An emerging disease causes regional population collapse of a common North American bat species. Science 329:679–682.
Froeschke, G., and S. Sommer. 2005. MHC class II DRB variability and parasite load in the striped mouse (*Rhabdomys pumilio*) in the southern Kalahari (vol 22, pg 1254, 2005). Molecular Biology and Evolution 22:1529.
Galindo, R. C., P. Ayoubi, V. Naranjo, C. Gortazar, K. M. Kocan, and J. de la Fuente. 2009. Gene expression profiles of European wild boar naturally infected with *Mycobacterium bovis*. Veterinary Immunology and Immunopathology 129:119–125.
Galli, L., A. Pereira, A. Márquez, and R. Mazzoni. 2006. Ranavirus detection by PCR in cultured tadpoles (*Rana catesbeiana* Shaw, 1802) from South America. Aquaculture 257:78–82.
Gariepy, T. D., R. Lindsay, N. Ogden, and T. R. Gregory. 2012. Identifying the last supper: utility of the DNA barcode library for bloodmeal identification in ticks. Molecular Ecology Resources 12:646–652.
Girard, J. M., D. M. Wagner, A. J. Vogler, C. Keys, C. J. Allender, L. C. Drickamer, and P. Keim. 2004. Differential plague-transmission dynamics determine *Yersinia pestis* population genetic structure on local, regional, and global scales. Proceedings of the National Academy of Sciences of the United States of America 101:8408–8413.
Grear, D. A., M. D. Samuel, K. T. Scribner, B. V. Weckworth, and J. A. Langenberg. 2010. Influence of genetic relatedness and spatial proximity on chronic wasting disease infection among female white-tailed deer. Journal of Applied Ecology 47:532–540.
Grenfell, B. T., O. G. Pybus, J. R. Gog, J. L. N. Wood, J. M. Daly, J. A. Mumford, and E. C. Holmes. 2004. Unifying the epidemiological and evolutionary dynamics of pathogens. Science 303:327–332.
Grice, E. A., and J. A. Segre. 2011. The skin microbiome. Nature Reviews Microbiology 9:244–253.
Hamer, G. L., U. D. Kitron, T. L. Goldberg, J. D. Brawn, S. R. Loss, M. O. Ruiz, D. B. Hayes, and E. D. Walker. 2009. Host selection by *Culex pipiens* mosquitoes and West Nile virus amplification. American Journal of Tropical Medicine and Hygiene 80:268–278.
Hansson, B., and L. Westerberg. 2002. On the correlation between heterozygosity and fitness in natural populations. Molecular Ecology 11:2467–2474.
Harrison, G. F., J. L. Scheirer, and V. R. Melanson. 2015. Development and validation of an arthropod maceration protocol for zoonotic pathogen detection in mosquitoes and fleas. Journal of Vector Ecology 40:83–89.
Harvell, C. D., C. E. Mitchell, J. R. Ward, S. Altizer, A. P. Dobson, R. S. Ostfeld, and M. D. Samuel. 2002. Ecology—climate warming and disease risks for terrestrial and marine biota. Science 296:2158–2162.
Hatta, M., P. Gao, P. Halfmann, and Y. Kawaoka. 2001. Molecular basis for high virulence of Hong Kong H5N1 influenza A viruses. Science 293:1840–1842.
Hedrick, P. W., T. J. Kim, and K. M. Parker. 2001. Parasite resistance and genetic variation in the endangered Gila topminnow. Animal Conservation 4:103–109.
Hellgren, O., S. Bensch, and B. Malmqvist. 2008. Bird hosts, blood parasites and their vectors—associations uncovered by molecular analyses of blackfly blood meals. Molecular Ecology 17:1605–1613.
Hellgren, O., J. Pérez-Tris, and S. Bensch. 2009. A jack-of-all-trades and still a master of some: prevalence and host range in avian malaria and related blood parasites. Ecology 90:2840–2849.
Hellgren, O., J. Waldenstrom, and S. Bensch. 2004. A new PCR assay for simultaneous studies of *Leucocytozoon*, *Plasmodium*, and *Haemoproteus* from avian blood. Journal of Parasitology 90:797–802.
Henaux, V., J. Parmley, C. Soos, and M. D. Samuel. 2013. Estimating transmission of avian influenza in wild birds from incomplete epizootic data: implications for surveillance and disease spread. Journal of Applied Ecology 50:223–231.
Hill, N. J., J. Y. Takekawa, J. T. Ackerman, K. A. Hobson, G. Herring, C. J. Cardona, J. A. Runstadler, and W. M. Boyce. 2012. Migration strategy affects avian influenza dynamics in mallards (*Anas platyrhynchos*). Molecular Ecology 21:5986–5999.
Hooper, L. V., D. R. Littman, and A. J. Macpherson. 2012. Interactions between the microbiota and the immune system. Science 336:1268–1273.
Hurtado, P. 2008. The potential impact of disease on the migratory structure of a partially migratory passerine population. Bulletin of Mathematical Biology 70:2264–2282.
James, T. Y., A. P. Litvintseva, R. Vilgalys, J. A. Morgan, J. W. Taylor, M. C. Fisher, L. Berger, C. Weldon, L. du Preez, and J. E. Longcore. 2009. Rapid global expansion of the fungal disease chytridiomycosis into declining and healthy amphibian populations. PLoS Pathogens 5: e1000458.
Johnson, C., J. Johnson, J. P. Vanderloo, D. Keane, J. M. Aiken, and D. McKenzie. 2006. Prion protein polymorphisms in white-tailed deer influence susceptibility to chronic wasting disease. Journal of General Virology 87:2109–2114.

Johnson, C. J., A. Herbst, C. Duque-Velasquez, J. P. Vanderloo, P. Bochsler, R. Chappell, and D. McKenzie. 2011. Prion protein polymorphisms affect chronic wasting disease progression. PLoS ONE 6:e17450.

Johnson, L. J. A. N., A. N. Miller, R. A. McCleery, R. McClanahan, J. A. Kath, S. Lueschow, and A. Porras-Alfaro. 2013. Psychrophilic and psychrotolerant fungi on bats and the presence of *Geomyces* spp. on bat wings prior to the arrival of white nose syndrome. Applied and Environmental Microbiology 79:5465–5471.

Jones, K. E., N. G. Patel, M. A. Levy, A. Storeygard, D. Balk, J. L. Gittleman, and P. Daszak. 2008*a*. Global trends in emerging infectious diseases. Nature 451:990–994.

Jones, M. E., A. Cockburn, R. Hamede, C. Hawkins, H. Hesterman, S. Lachish, D. Mann, H. McCallum, and D. Pemberton. 2008*b*. Life-history change in disease-ravaged Tasmanian devil populations. Proceedings of the National Academy of Sciences of the United States of America 105:10023–10027.

Joseph, M. B., J. R. Mihaljevic, A. L. Arellano, J. G. Kueneman, D. L. Preston, P. C. Cross, and P. T. J. Johnson. 2013. Taming wildlife disease: bridging the gap between science and management. Journal of Applied Ecology 50:702–712.

Kamath, P. L., J. T. Foster, K. P. Drees, G. Luikart, C. Quance, N. J. Anderson, P. R. Clarke, E. K. Cole, M. L. Drew, W. H. Edwards, J. C. Rhyan, J. J. Treanor, R. L. Wallen, P. J. White, S. Robbe-Austerman, and P. C. Cross. 2016. Genomics reveals historic and contemporary transmission dynamics of a bacterial disease among wildlife and livestock. Nature Communications 7:In press.

Kelly, A. C., N. E. Mateus-Pinilla, W. Brown, M. O. Ruiz, M. R. Douglas, M. E. Douglas, P. Shelton, T. Beissel, and J. Novakofski. 2014. Genetic assessment of environmental features that influence deer dispersal: implications for prion-infected populations. Population Ecology 56:327–340.

Kelly, A. C., N. E. Mateus-Pinilla, J. Diffendorfer, E. Jewell, M. O. Ruiz, J. Killefer, P. Shelton, T. Beissel, and J. Novakofski. 2008. Prion sequence polymorphisms and chronic wasting disease resistance in Illinois white-tailed deer (*Odocoileus virginianus*). Prion 2:28–36.

Kelly, A. C., N. E. Mateus-Pinilla, M. Douglas, M. Douglas, W. Brown, M. O. Ruiz, J. Killefer, P. Shelton, T. Beissel, and J. Novakofski. 2010. Utilizing disease surveillance to examine gene flow and dispersal in white-tailed deer. Journal of Applied Ecology 47:1189–1198.

Kenefic, L. J., T. Pearson, R. T. Okinaka, J. M. Schupp, D. M. Wagner, J. Ravel, A. R. Hoffmaster, C. P. Trim, W. K. Chung, J. A. Beaudry, J. T. Foster, J. I. Mead, and P. Keim. 2009. Pre-columbian origins for North American anthrax. PLoS ONE 4:e4813.

Kent, R. J. 2009. Molecular methods for arthropod bloodmeal identification and applications to ecological and vector-borne disease studies. Molecular Ecology Resources 9:4–18.

Knowles, S. C., M. J. Wood, R. Alves, T. A. Wilkin, S. Bensch, and B. C. Sheldon. 2011. Molecular epidemiology of malaria prevalence and parasitaemia in a wild bird population. Molecular Ecology 20:1062–1076.

Köser, C. U., M. J. Ellington, and S. J. Peacock. 2014. Whole-genome sequencing to control antimicrobial resistance. Trends in Genetics 30:401–407.

Lachish, S., H. McCallum, and M. Jones. 2009. Demography, disease and the devil: life-history changes in a disease-affected population of Tasmanian devils (*Sarcophilus harrisii*). Journal of Animal Ecology 78:427–436.

Lachish, S., K. J. Miller, A. Storfer, A. W. Goldizen, and M. E. Jones. 2011. Evidence that disease-induced population decline changes genetic structure and alters dispersal patterns in the Tasmanian devil. Heredity 106:172–182.

Lam, T. T. Y., H. S. Ip, E. Ghedin, D. E. Wentworth, R. A. Halpin, T. B. Stockwell, D. J. Spiro, R. J. Dusek, J. B. Bortner, J. Hoskins, B. D. Bales, D. R. Yparraguirre, and E. C. Holmes. 2012. Migratory flyway and geographical distance are barriers to the gene flow of influenza virus among North American birds. Ecology Letters 15:24–33.

Lane-deGraaf, K. E., S. J. Amish, F. Gardipee, A. Jolles, G. Luikart, and V. O. Ezenwa. 2015. Signatures of natural and unnatural selection: evidence from an immune system gene in African buffalo. Conservation Genetics 16:289–300.

Lang, K. R., and J. A. Blanchong. 2012. Population genetic structure of white-tailed deer: understanding risk of chronic wasting disease spread. Journal of Wildlife Management 76:832–840.

Langwig, K. E., J. R. Hoyt, K. L. Parise, J. Kath, D. Kirk, W. F. Frick, J. T. Foster, and A. M. Kilpatrick. 2015. Invasion dynamics of white-nose syndrome fungus, Midwestern United States, 2012–2014. Emerging Infectious Diseases 21:1023–1026.

Latorre-Margalef, N., V. Grosbois, J. Wahlgren, V. J. Munster, C. Tolf, R. A. M. Fouchier, A. D. M. E. Osterhaus, B. Olsen, and J. Waldenstrom. 2013. Heterosubtypic immunity to influenza A virus infections in mallards may explain existence of multiple virus subtypes. Plos Pathogens 9: e1003443.

Latorre-Margalef, N., C. Tolf, V. Grosbois, A. Avril, D. Bengtsson, M. Wille, A. D. M. E. Osterhaus, R. A. M. Fouchier, B. Olsen, and J. Waldenstrom. 2014. Long-term variation in influenza A virus prevalence and subtype diversity in migratory mallards in northern Europe. Proceedings of the Royal Society B: Biological Sciences 281:20140098.

le Roex, N., P. D. van Helden, A. P. Koets, and E. G. Hoal. 2013. Bovine TB in livestock and wildlife: what's in the genes? Physiological Genomics 45:631–637.

Lee, J. S., S. N. Bevins, L. E. K. Serieys, W. Vickers, K. A. Logan, M. Aldredge, E. E. Boydston, L. M. Lyren, R. McBride, M. Roelke-Parker, J. Pecon-Slattery, J. L. Troyer, S. P. Riley, W. M. Boyce, K. R. Crooks, and S. VandeWoude. 2014. Evolution of puma lentivirus in bobcats (*Lynx rufus*) and mountain lions (*Puma concolor*) in North America. Journal of Virology 88:7727–7737.

Lee, J. S., E. W. Ruell, E. E. Boydston, L. M. Lyren, R. S. Alonso, J. L. Troyer, K. R. Crooks, and S. VandeWoude. 2012. Gene flow and pathogen transmission among bobcats (*Lynx rufus*) in a fragmented urban landscape. Molecular Ecology 21:1617–1631.

Lee, K. E., J. M. Seddon, S. W. Corley, W. A. H. Ellis, S. D. Johnston, D. L. de Villiers, H. J. Preece, and F. N. Carrick. 2010. Genetic variation and structuring in the threatened koala populations of Southeast Queensland. Conservation Genetics 11:2091–2103.

Leiser, O. P., J. L. Corn, B. S. Schmit, P. S. Keim, and J. T. Foster. 2013. Feral swine brucellosis in the United States and prospective genomic techniques for disease epidemiology. Veterinary Microbiology 166:1–10.

Lipkin, W. I. 2013. The changing face of pathogen discovery and surveillance. Nature Reviews Microbiology 11:133–141.

Liu, D., editor. 2011. Molecular detection of human fungal pathogens. CRC Press, Boca Raton, Florida, USA.

Loiseau, C., R. Zoorob, S. Garnier, J. Birard, P. Federici, R. Julliard, and G. Sorci. 2008. Antagonistic effects of a MHC class I allele on malaria-infected house sparrows. Ecology Letters 11:258–265.

Loiseau, C., R. Zoorob, A. Robert, O. Chastel, R. Julliard, and G. Sorci. 2011. *Plasmodium relictum* infection and MHC diversity in the house sparrow (*Passer domesticus*). Proceedings of the Royal Society B-Biological Sciences 278:1264–1272.

Lorch, J. M., A. Gargas, C. U. Meteyer, B. M. Berlowski-Zier, D. E. Green, V. Shearn-Bochsler, N. J. Thomas, and D. S. Blehert. 2010. Rapid polymerase chain reaction diagnosis of white-nose syndrome in bats. Journal of Veterinary Diagnostic Investigation 22:224–230.

Luikart, G., P. R. England, D. Tallmon, S. Jordan, and P. Taberlet. 2003. The power and promise of population genomics: from genotyping to genome typing. Nature Reviews Genetics 4:981–994.

Magle, S. B., M. D. Samuel, T. R. Van Deelen, S. J. Robinson, and N. E. Mathews. 2013. Evaluating spatial overlap and relatedness of white-tailed deer in a chronic wasting disease management zone. PLos ONE 8:e56568.

Manel, S., and R. Holderegger. 2013. Ten years of landscape genetics. Trends in Ecology & Evolution 28:614–621.

Manel, S., O. E. Gaggiotti, and R. S. Waples. 2005. Assignment methods: matching biological questions with appropriate techniques. Trends in Ecology & Evolution 20:136–142.

Manel, S., M. K. Schwartz, G. Luikart, and P. Taberlet. 2003. Landscape genetics: combining landscape ecology and population genetics. Trends in Ecology & Evolution 18:189–197.

Matsumoto, T., M. D. Samuel, T. Bollinger, M. Pybus, and D. W. Coltman. 2013. Association mapping of genetic risk factors for chronic wasting disease in wild deer. Evolutionary Applications 6:340–352.

McCallum, H. 2008. Tasmanian devil facial tumour disease: lessons for conservation biology. Trends in Ecology & Evolution 23:631–637.

McCallum, H., N. Barlow, and J. Hone. 2001. How should pathogen transmission be modelled? Trends in Ecology & Evolution 16:295–300.

McCarthy, A. J., M. A. Shaw, P. D. Jepson, S. M. J. M. Brasseur, P. J. H. Reijnders, and S. J. Goodman. 2011. Variation in European harbour seal

immune response genes and susceptibility to phocine distemper virus (PDV). Infection, Genetics and Evolution 11:1616–1623.
Miller, R. R., V. Montoya, J. L. Gardy, D. M. Patrick, and P. Tang. 2013. Metagenomics for pathogen detection in public health. Genome Medicine 5:81.
Molaei, G., T. G. Andreadis, P. M. Armstrong, J. F. Anderson, and C. R. Vossbrinck. 2006. Host feeding patterns of *Culex* mosquitoes and West Nile virus transmission, northeastern United States. Emerging Infectious Diseases 12:468–474.
Morand, S., F. Beaudeau, and J. Cabaret, editors. 2012. New frontiers of molecular epidemiology of infectious diseases. Springer, Dordrecht, Netherlands.
Morelli, G., Y. Song, C. J. Mazzoni, M. Eppinger, P. Roumagnac, D. M. Wagner, M. Feldkamp, B. Kusecek, A. J. Vogler, Y. Li, Y. Cui, N. R. Thomson, T. Jombart, R. Leblois, P. Lichtner, L. Rahalison, J. M. Petersen, F. Balloux, P. Keim, T. Wirth, J. Ravel, R. Yang, E. Carniel, and M. Achtman. 2010. *Yersinia pestis* genome sequencing identifies patterns of global phylogenetic diversity. Nature Genetics 42:1140–1143.
Muller, L. K., J. M. Lorch, D. L. Lindner, M. O'Connor, A. Gargas, and D. S. Blehert. 2013. Bat white-nose syndrome: a real-time TaqMan polymerase chain reaction test targeting the intergenic spacer region of Geomyces destructans. Mycologia 105:253–259.
Naccache, S. N., S. Federman, N. Veeraraghavan, M. Zaharia, D. Lee, E. Samayoa, J. Bouquet, A. L. Greninger, K. C. Luk, B. Enge, D. A. Wadford, S. L. Messenger, G. L. Genrich, K. Pellegrino, G. Grard, E. Leroy, B. S. Schneider, J. N. Fair, M. A. Martinez, P. Isa, J. A. Crump, J. L. DeRisi, T. Sittler, J. Hackett Jr., S. Miller, and C. Y. Chiu. 2014. A cloud-compatible bioinformatics pipeline for ultrarapid pathogen identification from next-generation sequencing of clinical samples. Genome Research 24:1180–1192.
Niskanen, A. K., L. J. Kennedy, M. Ruokonen, I. Kojola, H. Lohi, M. Isomursu, E. Jansson, T. Pyhajarvi, and J. Aspi. 2014. Balancing selection and heterozygote advantage in major histocompatibility complex loci of the bottlenecked Finnish wolf population. Molecular Ecology 23:875–889.
O'Rourke, K. I., T. E. Besser, M. W. Miller, T. F. Cline, T. R. Spraker, A. L. Jenny, M. A. Wild, G. L. Zebarth, and E. S. Williams. 1999. PrP genotypes of captive and free-ranging Rocky Mountain elk (*Cervus elaphus nelsoni*) with chronic wasting disease. Journal of General Virology 80:2765–2769.
Olden, J. D., N. L. Poff, M. R. Douglas, M. E. Douglas, and K. D. Fausch. 2004. Ecological and evolutionary consequences of biotic homogenization. Trends in Ecology & Evolution 19:18–24.
Oliver, M. K., S. Telfer, and S. B. Piertney. 2009. Major histocompatibility complex (MHC) heterozygote superiority to natural multi-parasite infections in the water vole (*Arvicola terrestris*). Proceedings of the Royal Society B-Biological Sciences 276:1119–1128.
Orme, I. 2004. Adaptive immunity to mycobacteria. Current Opinion in Microbiology 7:58–61.
Osborne, A. J., J. Pearson, S. S. Negro, B. L. Chilvers, M. A. Kennedy, and N. J. Gemmell. 2015. Heterozygote advantage at MHC DRB may influence response to infectious disease epizootics. Molecular Ecology 24:1419–1432.
Ostfeld, R. S., G. E. Glass, and F. Keesing. 2005. Spatial epidemiology: an emerging (or re-emerging) discipline. Trends in Ecology & Evolution 20:328–336.
Panzera, Y., N. Sarute, G. Iraola, M. Hernandez, and R. Perez. 2015. Molecular phylogeography of canine distemper virus: geographic origin and global spreading. Molecular Phylogenetics and Evolution 92:147–154.
Paquette, S. R., B. Talbot, D. Garant, J. Mainguy, and F. Pelletier. 2014. Modelling the dispersal of the two main hosts of the raccoon rabies variant in heterogeneous environments with landscape genetics. Evolutionary Applications 7:734–749.
Pasick, J. 2008. Advances in the molecular based techniques for the diagnosis and characterization of avian influenza virus infections. Transboundary and Emerging Diseases 55:329–338.
Pecon-Slattery, J., C. L. McCracken, J. L. Troyer, S. VandeWoude, M. Roelke, K. Soundgeroth, C. Winterbach, H. Winterbach, and S. J. O'Brien. 2008. Genomic organization, sequence divergence, and recombination of feline immunodeficiency virus from lions in the wild. BMC Genomics 9:66.
Pedersen, A. B., and S. A. Babayan. 2011. Wild immunology. Molecular Ecology 20:872–880.
Perucchini, M., K. Griffin, M. W. Miller, and W. Goldmann. 2008. PrP genotypes of free-ranging wapiti (*Cervus elaphus nelsoni)* with chronic wasting disease. Journal of General Virology 89:1324–1328.
Porter, K. R., P. L. Summers, D. Dubois, B. Puri, W. Nelson, E. Henchal, J. J. Oprandy, and C. G. Hayes. 1993. Detection of West Nile virus by the polymerase chain reaction and analysis of nucleotide sequence variation. American Journal of Tropical Medicine and Hygiene 48:440–446.
Raberg, L., D. Sim, and A. F. Read. 2007. Disentangling genetic variation for resistance and tolerance to infectious diseases in animals. Science 318:812–814.
Rampini, S. K., G. V. Bloemberg, P. M. Keller, A. C. Büchler, G. Dollenmaier, R. F. Speck, and E. C. Böttger. 2011. Broad-range 16S rRNA gene polymerase chain reaction for diagnosis of culture-negative bacterial infections. Clinical Infectious Diseases 53:1245–1251.
Rawat, A., D. M. Engelthaler, E. M. Driebe, P. Keim, and J. T. Foster. 2014. MetaGeniE: characterizing human clinical samples using deep metagenomic sequencing. PLoS ONE 9:e110915.
Remais, J. V., N. Xiao, A. Akullian, D. Qiu, and D. Blair. 2011. Genetic assignment methods for gaining insight into the management of infectious disease by understanding pathogen, vector, and host movement. PLoS Pathogens 7:e1002013.
Robinson, R. A., B. Lawson, M. P. Toms, K. M. Peck, J. K. Kirkwood, J. Chantrey, I. R. Clatworthy, A. D. Evans, L. A. Hughes, O. C. Hutchinson, S. K. John, T. W. Pennycott, M. W. Perkins, P. S. Rowley, V. R. Simpson, K. M. Tyler, and A. A. Cunningham. 2010. Emerging infectious disease leads to rapid population declines of common British birds. PLoS ONE 5:e12215.
Robinson, S. J., M. D. Samuel, C. J. Johnson, M. Adams, and D. I. McKenzie. 2012*a*. Emerging prion disease drives host selection in a wildlife population. Ecological Applications 22:1050–1059.
Robinson, S. J., M. D. Samuel, D. L. Lopez, and P. Shelton. 2012*b*. The walk is never random: subtle landscape effects shape gene flow in a continuous white-tailed deer population in the Midwestern United States. Molecular Ecology 21:4190–4205.
Robinson, S. J., M. D. Samuel, K. I. O'Rourke, and C. J. Johnson. 2012*c*. The role of genetics in chronic wasting disease of North American cervids. Prion 6:153–162.
Robinson, S. J., M. D. Samuel, R. E. Rolley, and P. Shelton. 2013. Using landscape epidemiological models to understand the distribution of chronic wasting disease in the Midwestern USA. Landscape Ecology 28:1923–1935.
Rogers, K. G., S. J. Robinson, M. D. Samuel, and D. A. Grear. 2011. Diversity and distribution of white-tailed deer mtDNA linages in chronic wasting disease (CWD) outbreak areas in southern Wisconsin, USA. Journal of Toxicology and Environmental Health—Part A—Current Issues 74:1521–1535.
Rolfs, A., I. Schuller, U. Finckh, and I. Weber-Rolfs. 1992. PCR: clinical diagnostics and research. Springer-Verlag, Berlin, Germany.
Rosenblum, E. B., T. Y. James, K. R. Zamudio, T. J. Poorten, D. Ilut, D. Rodriguez, J. M. Eastman, K. Richards-Hrdlicka, S. Joneson, T. S. Jenkinson, J. E. Longcore, G. Parra Olea, L. F. Toledo, M. L. Arellano, E. M. Medina, S. Restrepo, S. V. Flechas, L. Berger, C. J. Briggs, and J. E. Stajich. 2013. Complex history of the amphibian-killing chytrid fungus revealed with genome resequencing data. Proceedings of the National Academy of Sciences of the United States of America 110:9385–9390.
Roug, A., C. Geoghegan, E. Wellington, W. A. Miller, E. Travis, D. Porter, D. Cooper, D. L. Clifford, J. A. K. Mazet, and S. Parsons. 2014. Utility of a fecal real-time PCR protocol for detection of *Mycobacterium bovis* infection in African buffalo (*Syncerus caffer*). Journal of Wildlife Diseases 50:140–142.
Russell, C. A., L. A. Real, and D. L. Smith. 2006. Spatial control of rabies on heterogeneous landscapes. PLoS ONE 1:e27.
Sahl, J., J. Schupp, D. Rasko, R. Colman, J. Foster, and P. Keim. 2015. Phylogenetically typing bacterial strains from partial SNP genotypes observed from direct sequencing of clinical specimen metagenomic data. Genome Medicine 7:52.
Samuel, M. D., B. L. Woodworth, C. T. Atkinson, P. J. Hart, and D. A. LaPointe. 2015. Avian malaria in Hawaiian forest birds: infection and population impacts across species and elevations. Ecosphere 6:104.

Savage, A. E., and K. R. Zamudio. 2011. MHC genotypes associate with resistance to a frog-killing fungus. Proceedings of the National Academy of Sciences of the United States of America 108:16705–16710.
Schwensow, N., J. Fietz, K. H. Dausmann, and S. Sommer. 2007. Neutral versus adaptive genetic variation in parasite resistance: importance of major histocompatibility complex supertypes in a free-ranging primate. Heredity 99:265–277.
Segura, V., B. J. Vilhjalmsson, A. Platt, A. Korte, U. Seren, Q. Long, and M. Nordborg. 2012. An efficient multi-locus mixed-model approach for genome-wide association studies in structured populations. Nature Genetics 44:825–830.
Sepil, I., S. Lachish, A. E. Hinks, and B. C. Sheldon. 2013. Mhc supertypes confer both qualitative and quantitative resistance to avian malaria infections in a wild bird population. Proceedings of the Royal Society B-Biological Sciences 280:0130134.
Serieys, L. E. K., A. Lea, J. P. Pollinger, S. P. D. Riley, and R. K. Wayne. 2015. Disease and freeways drive genetic change in urban bobcat populations. Evolutionary Applications 8:75–92.
Shafer, A. B. A., J. M. Northrup, M. Wikelski, G. Wittemyer, and J. B. W. Wolf. 2016. Forecasting ecological genomics: high-tech animal instrumentation meets high-throughput sequencing. PLoS Biology 14: e1002350.
Shafer, A. B., J. B. Wolf, P. C. Alves, L. Bergstrom, M. W. Bruford, I. Brannstrom, G. Colling, L. Dalen, L. De Meester, R. Ekblom, K. D. Fawcett, S. Fior, M. Hajibabaei, J. A. Hill, A. R. Hoezel, J. Hoglund, E. L. Jensen, J. Krause, T. N. Kristensen, M. Krutzen, J. K. McKay, A. J. Norman, R. Ogden, E. M. Osterling, N. J. Ouborg, J. Piccolo, D. Popovic, C. R. Primmer, F. A. Reed, M. Roumet, J. Salmona, T. Schenekar, M. K. Schwartz, G. Segelbacher, H. Senn, J. Thaulow, M. Valtonen, A. Veale, P. Vergeer, N. Vijay, C. Vila, M. Weissensteiner, L. Wennerstrom, C. W. Wheat, and P. Zielinski. 2015. Genomics and the challenging translation into conservation practice. Trends in Ecology & Evolution 30:78–87.
Siddle, H. V., A. Kreiss, M. D. B. Eldridge, E. Noonan, C. J. Clarke, S. Pyecroft, G. M. Woods, and K. Belov. 2007. Transmission of a fatal clonal tumor by biting occurs due to depleted MHC diversity in a threatened carnivorous marsupial. Proceedings of the National Academy of Sciences of the United States of America 104:16221–16226.
Silvy, N. J., editor. 2012. The wildlife techniques manual: volume 1: research. 7th edition. Johns Hopkins University Press, Baltimore, Maryland, USA.
Skerratt, L. F., L. Berger, R. Speare, S. Cashins, K. R. McDonald, A. D. Phillott, H. B. Hines, and N. Kenyon. 2007. Spread of chytridiomycosis has caused the rapid global decline and extinction of frogs. Ecohealth 4:125–134.
Sorci, G. 2013. Immunity, resistance and tolerance in bird-parasite interactions. Parasite Immunology 35:350–361.
Spielman, D., B. W. Brook, D. A. Briscoe, and R. Frankham. 2004. Does inbreeding and loss of genetic diversity decrease disease resistance? Conservation Genetics 5:439–448.
Steiner, C. C., A. S. Putnam, P. E. A. Hoeck, and O. A. Ryder. 2013. Conservation genomics of threatened animal species. Annual Review of Animal Biosciences 1:261–281.
Talbot, B., D. Garant, S. R. Paquette, J. Mainguy, and F. Pelletier. 2013. Genetic structure and diversity among rabid and nonrabid raccoons. Ecoscience 20:345–351.
Teacher, A. G. F., T. W. J. Garner, and R. A. Nichols. 2009. Population genetic patterns suggest a behavioural change in wild common frogs (*Rana temporaria*) following disease outbreaks (*Ranavirus*). Molecular Ecology 18:3163–3172.
Trudeau, K. M., H. B. Britten, and M. Restani. 2004. Sylvatic plague reduces genetic variability in black-tailed prairie dogs. Journal of Wildlife Diseases 40:205–211.
Tschirren, B. 2015. *Borrelia burgdorferi* sensu lato infection pressure shapes innate immune gene evolution in natural rodent populations across Europe. Biology Letters 11:20150263.
Turner, A. K., M. Begon, J. A. Jackson, and S. Paterson. 2012. Evidence for selection at cytokine loci in a natural population of field voles (*Microtus agrestis*). Molecular Ecology 21:1632–1646.
Vali, U., A. Einarsson, L. Waits, and H. Ellegren. 2008. To what extent do microsatellite markers reflect genome-wide genetic diversity in natural populations? Molecular Ecology 17:3808–3817.
van Riper III, C., S. G. van Riper, M. L. Goff, and M. Laird. 1986. The epizootiology and ecological significance of malaria in Hawaiian land birds. Ecological Monographs 56:327–344.
Vander Wal, E., I. Edye, P. C. Paquet, D. W. Coltman, E. Bayne, R. K. Brook, and J. A. Andres. 2013. Juxtaposition between host population structures: implications for disease transmission in a sympatric cervid community. Evolutionary Applications 6:1001–1011.
VanderWaal, K. L., E. R. Atwill, L. A. Isbell, and B. McCowan. 2014. Quantifying microbe transmission networks for wild and domestic ungulates in Kenya. Biological Conservation 169:136–146.
Vogler, A. J., D. Birdsell, L. B. Price, J. R. Bowers, S. M. Beckstrom-Sternberg, R. K. Auerbach, J. S. Beckstrom-Sternberg, A. Johansson, A. Clare, J. L. Buchhagen, J. M. Petersen, T. Pearson, J. Vaissaire, M. P. Dempsey, P. Foxall, D. M. Engelthaler, D. M. Wagner, and P. Keim. 2009. Phylogeography of *Francisella tularensis*: global expansion of a highly fit clone. Journal of Bacteriology 191:2474–2484.
Volz, E. M., K. Koelle, and T. Bedford. 2013. Viral phylodynamics. PLoS Computational Biology 9:e1002947.
Westerdahl, H., M. Asghar, D. Hasselquist, and S. Bensch. 2012. Quantitative disease resistance: to better understand parasite-mediated selection on major histocompatibility complex. Proceedings of the Royal Society B-Biological Sciences 279:577–584.
Westermann, A. J., S. A. Gorski, and J. Vogel. 2012. Dual RNA-seq of pathogen and host. Nature Reviews Microbiology 10:618–630.
Wheeler, D. C., L. A. Waller, and R. Biek. 2010. Spatial analysis of feline immunodeficiency virus infection in cougars. Spatial and Spatio-temporal Epidemiology 1:151–161.
White, L. A., J. D. Forester, and M. E. Craft. 2016. Using contact networks to explore mechanisms of parasite transmission in wildlife. Biological Reviews 91:In press.
Whiteman, N. K., K. D. Matson, J. L. Bollmer, and P. G. Parker. 2006. Disease ecology in the Galápagos hawk (*Buteo galapagoensis*): host genetic diversity, parasite load and natural antibodies. Proceedings of the Royal Society B: Biological Sciences 273:797–804.
Wilson, G. A., S. M. Nakada, T. K. Bollinger, M. J. Pybus, E. H. Merrill, and D. W. Coltman. 2009. Polymorphisms at the PRNP gene influence susceptibility to chronic wasting disease in two species of deer (*Odocoileus* spp.) in western Canada. Journal of Toxicology and Environmental Health —Part A—Current Issues 72:1025–1029.
Woodworth, B. L., C. T. Atkinson, D. A. LaPointe, P. J. Hart, C. S. Spiegel, E. J. Tweed, C. Henneman, J. LeBrun, T. Denette, R. DeMots, K. L. Kozar, D. Triglia, D. Lease, A. Gregor, T. Smith, and D. Duffy. 2005. Host population persistence in the face of introduced vector-borne diseases: Hawaii amakihi and avian malaria. Proceedings of the National Academy of Sciences of the United States of America 102:1531–1536.
Worley, K., J. Collet, L. G. Spurgin, C. Cornwallis, T. Pizzari, and D. S. Richardson. 2010. MHC heterozygosity and survival in red junglefowl. Molecular Ecology 19:3064–3075.
Yamamoto, Y. 2002. PCR in diagnosis of infection: detection of bacteria in cerebrospinal fluids. Clinical and Diagnostic Laboratory Immunology 9:508–514.

Associate Editor: Zachary Olson.

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article at the publisher's website.