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Population Genomics for Insect Conservation

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Abstract

Insects constitute vital components of ecosystems. There is alarming evidence for global declines in insect species diversity, abundance, and biomass caused by anthropogenic drivers such as habitat degradation or loss, agricultural practices, climate change, and environmental pollution. This raises important concerns about human food security and ecosystem functionality and calls for more research to assess insect population trends and identify threatened species and the causes of declines to inform conservation strategies. Analysis of genetic diversity is a powerful tool to address these goals, but so far animal conservation genetics research has focused strongly on endangered vertebrates, devoting less attention to invertebrates, such as insects, that constitute most biodiversity. Insects' shorter generation times and larger population sizes likely necessitate different analytical methods and management strategies. The availability of high-quality reference genome assemblies enables population genomics to address several key issues. These include precise inference of past demographic fluctuations and recent declines, measurement of genetic load levels, delineation of evolutionarily significant units and cryptic species, and analysis of genetic adaptation to stressors. This enables identification of populations that are particularly vulnerable to future threats, considering their potential to adapt and evolve. We review the application of population genomics to insect conservation and the outlook for averting insect declines.

1. INTRODUCTION

The advent of next-generation sequencing technologies enabled population genomics to be applied to any non-model organism, leading to the birth of conservation genomics (1–5). The ability to perform genome-scale analyses allows researchers to address questions that were intractable previously using small numbers of neutral genetic markers. These include detailed inference of demographic history, estimation of genetic load, and identification of genes involved in fitness-related traits.

So far, conservation genomics has been applied mainly to endangered terrestrial vertebrates. However, these represent only a small fraction of Earth's biodiversity. Several reports have raised the possibility that insects are in sharp decline, in both abundance and species diversity (6–11). It is therefore important to evaluate progress in insect population genomics and to consider how this field can contribute to insect conservation. We review evidence for insect declines and their drivers. We then consider how population genomics can be applied to insect conservation, taking into account their inherent differences from vertebrates, which include a broad diversity in life histories and ecological strategies, short generation times, and often huge population sizes.

2. INSECT DECLINES: EVIDENCE AND DRIVERS

2.1. Are We Experiencing an Insect Apocalypse?

The impact of human activity on the planet is responsible for huge global loss of biodiversity due to habitat destruction and degradation, harvesting of natural populations, climate change, release of toxic chemicals, and expansion of invasive species. In 2019, a United Nations report (12) estimated that approximately one million animal and plant species are threatened with extinction. Species abundance is declining globally, with a reduction of at least 20% of native species on most terrestrial habitats estimated in the last decade. In marine habitats, a substantial proportion of corals, sharks, and mammals are also threatened with extinction, as are more than 40% of amphibian species (12).

Of the ~11 million animal species estimated to live on Earth, ~5.5 million are insects, of which ~1 million have been cataloged (13, 14). Insects are vital components of ecosystems. They are important sources of food for a huge range of vertebrates and play a vital role in a wide range of ecosystem functions, most notably decomposition and plant pollination. Several recent reports have highlighted insect declines, indicating that losses could exceed even those found in other taxonomic groups (6–11). Large numbers of species may therefore face extinction before they are known. Insect declines result in knock-on effects throughout food webs and can lead to extinctions (15). However, compared to those in well-studied vertebrate species such as birds and mammals, comprehensive studies of the conservation status of insects are still lacking.

Reports of insect declines focus mainly on population trends, finding declines in total biomass, abundance, range size, and species richness (6–11). One of the most comprehensive studies, focusing on flying insects in 63 sites in northwestern Germany, indicated a 75% reduction in biomass over 27 years (8). Long-term distribution data from the United Kingdom show a large fraction of insects have experienced range contraction (11). However, reported population trends are heterogeneous across insect taxonomic groups, habitats, and geographical regions, with most studies coming from Western and Northern Europe and the United States. There are comparatively few data from the tropics (6), although one key long-term study found evidence for insect biomass declines in a tropical forest (16).

Historical records of abundance are richest for butterflies, moths, and bees (17–20). In the United Kingdom, at least 70% of butterfly species are estimated to have declined in occurrence over the past four decades, and five species have disappeared (21). A study in a Swedish nature

reserve indicated a 45% loss of butterfly species over a 50-year period (22). A 31% decrease over 35 years was reported for macromoths in the United Kingdom (23). Massive winter losses of managed honeybee colonies in Europe and the United States in 2007 initially prompted alarm over loss of bees (24). Bumblebee declines have also been reported in North America and Europe, particularly in the southern margins of distributions that are affected most strongly by climate change (25). Trends are far more poorly documented for the vast majority of (mainly solitary) bee species. A historical study of bees from the northeastern United States over 140 years found no evidence for substantial declines among 187 native bee species, although large shifts in relative abundances were observed (26). In the United Kingdom, a 32% decline in abundance in solitary bees has been reported, particularly affecting rarer species (17).

Despite clear evidence for dramatic insect declines from local surveys, we are still far from a global understanding of insect population trends (6). A highly publicized meta-analysis proposed that global insect biomass was decreasing at an annual rate of 2.5% (7). However, this study has been criticized for heavy reliance on surveys conducted in the vicinity of regions with high human population density and habitat loss due to agriculture, and whether these data can be extrapolated to the rest of the world is unclear (27, 28). Another recent meta-analysis extrapolated from the available European and North American data to infer an annual 1% loss of terrestrial insects but a 1% increase in aquatic insects (10). Similarly, an analysis of population trends from time-series data for more than 6,000 European animal and plant species, including insects, found that freshwater insect species had increased in diversity, whereas terrestrial insects had strongly declined (29). An analysis of insect population trends across US research sites found no evidence of continent-wide decline (30), although it has been argued that this study does not account for complex sampling histories at field sites (31).

Currently available studies of insect population trends are strongly biased geographically and often focused on specific taxonomic groups. Assessments generally focus on habitats that have not changed over time (6, 32), and sampling sites affected by changes in land use due to agriculture or urbanization are often excluded from analyses of insect population trends (30, 31). Considering that habitat loss is a primary cause of insect declines, this could lead to the extent of declines being underestimated. On the other hand, many time-series studies have been performed in densely populated countries and may still be negatively impacted by human activity in areas surrounding the study sites even if the sites themselves are unaltered (6–8). Studies that do not find notable changes in diversity or abundance are also likely underreported. There is therefore great uncertainty in measures of the extent of insect declines and an urgent need to develop and implement new methods to generate an accurate account of population trends across taxonomic groups and geographical regions.

2.2. Drivers of Insect Declines

Most studies of insect declines point to habitat loss and insecticide use as the major drivers (6, 7). However, many other factors likely play an important role, notably climate change, parasites and pathogens, invasive species, and other pollutants. Specific drivers differ according to geography, and their effects vary greatly among species (33). Climate change will likely become an increasingly important driver if it continues at current rates.

The main causes of habitat loss are deforestation and destruction of natural habitats, usually due to expansion of agriculture and urban development. The ongoing clearing of tropical forests on a massive scale is causing a huge loss of insect biodiversity (34, 35). Most insect biodiversity is found in tropical forests, but most tropical insect species are still undescribed, and the extent of these losses is therefore poorly understood (6, 14). Forest cover in Europe has also declined drastically during the last 12,000 years (36) due to historic deforestation (37, 38). However, not all

species are affected negatively by these changes, and the creation of parks and meadows may cause some pollinators, including certain bumblebee species, to increase in numbers (39). The most important factor associated with insect losses overall is likely the expansion of modern intensive farming practices, which entail planting extensive monocultures, leading to reduced diversity in pollinators and other terrestrial insects and providing conditions for agricultural pests to thrive (7, 40).

A major additional threat connected to modern agriculture is posed by pesticides, which are considered the second major driver of insect declines (6, 7). Much attention has focused on neonicotinoid pesticides, which have severe negative effects on the growth and survival of artificially exposed bee colonies (41–43). In addition, areas in the vicinity of crops treated with a neonicotinoid pesticide contain fewer solitary bees and reduced growth of bumblebee colonies (44). Butterfly declines in both the United Kingdom and United States have been linked to neonicotinoids (6). The massive declines in insect biomass found in Germany (8) could plausibly be linked to pesticide usage, as almost all of the sites investigated are near agricultural land (6). Although pesticides directly impact insects, other agricultural chemicals released into the environment also have substantial effects. Herbicides reduce floral diversity and lead to declines in insects associated with wild plants (45). Fertilizers cause shifts in floral abundance, which may specifically threaten rare specialist pollinator species (40) and also leads to decreases in insect diversity in general (46). The effects of these agrochemicals on aquatic insects could also be substantial due to direct toxicity or eutrophication of water (7).

Climate change is likely to be an increasingly large threat to insect biodiversity and abundance in future due to rising temperatures and more extreme weather events. A study of arthropod abundance in a tropical forest found evidence for declines driven by rising temperatures (16). In Europe and North America, the southern ranges of butterflies and bumblebees are shrinking, probably due to the warming climate (25, 47, 48). Based on current distributions, the ranges of most bumblebees in Europe are predicted to shrink significantly due to climate change (49). Greater occurrence of extreme weather events is an important driver of losses of bumblebees from the southern limits of their ranges (47) and likely affects other insects. However, indirect effects of climate change, such as increased incidence of drought and fires, may have more significant effects via changes in flora and their associated insect herbivores (50). Global warming can also potentially disrupt ecological interactions such as plant–pollinator associations (51, 52) and be a driving force for expansions of pests (53). For example, a desert locust outbreak in East Africa in 2019–2020 was likely linked to anthropogenic climate change due to an extreme rainy season (54).

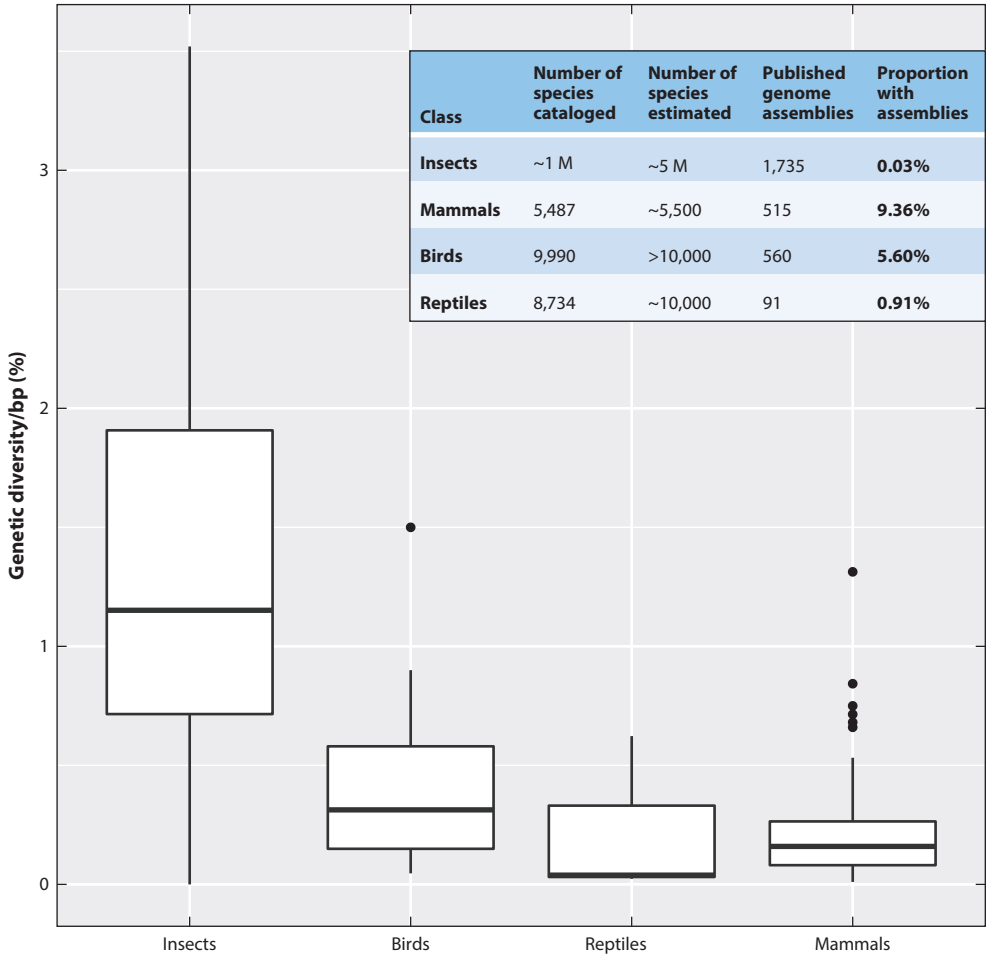
Human-mediated movement of animals and plants can facilitate biological invasions, which can negatively impact insect biodiversity (55). For example, invasive ants can outcompete or feed on native arthropods and can have severe ecological impacts (56). In South America, the bumblebees *Bombus terrestris* and *Bombus ruderatus* were introduced and became invasive, which is now causing a decline in the local Patagonian bumblebee *Bombus dahlbomii* population due to both competition and the introduction of a new parasite (59). Similarly, the invasive harlequin ladybird, *Harmonia axyridis*, carries parasitic microsporidia, which do not harm it but are lethal to native ladybird species in North America and Europe (60). There is evidence that viruses in managed bee colonies can spill over into wild bee species (57, 58). Expansions of vertebrates such as cattle, pigs, and rabbits can also damage insect diversity via increased consumption of host plants.

Population genomics could play an important role in assessing insect population trends and uncovering the drivers of biodiversity loss, which can inform strategies to mitigate their effects. Potential analyses include demographic modeling to analyze changes in population size in response to drivers and identification of adaptive genetic variation that enables prediction of how populations are expected to adapt and evolve in response to these drivers in future.

3. IMPORTANCE OF NEUTRAL GENETIC VARIATION

So far, conservation genetics research has focused on endangered terrestrial vertebrates, which are often found in small, fragmented populations with impoverished genetic variation. However, this may often not be the case for endangered insect species. Several intrinsic features of insects could indicate that different research goals and management strategies are often more appropriate for their conservation. In particular, insects generally have much larger population sizes, shorter generation times, and higher levels of heterozygosity than terrestrial vertebrates (**Figure 1**), requiring careful evaluation of the significance of levels of neutral genetic variation for insect conservation (61, 62).

An important goal in conservation genetics is estimation of effective population size (N_e), typically by assaying levels of genetic variation at neutral markers (63). N_e is defined as the size of a theoretical ideal population with the same degree of genetic drift as the study population.



Class	Number of species cataloged	Number of species estimated	Published genome assemblies	Proportion with assemblies
Insects	~1 M	~5 M	1,735	**0.03%**
Mammals	5,487	~5,500	515	**9.36%**
Birds	9,990	>10,000	560	**5.60%**
Reptiles	8,734	~10,000	91	**0.91%**

Figure 1

Levels of neutral genetic variation per base pair (bp) in insects (110 species), mammals (56 species), birds (18 species), and reptiles (3 species); data taken from Reference 62. Inset shows number of cataloged and estimated total number of extant species in each class; data taken from Reference 13 and number of genome assemblies taken from the National Center for Biotechnology Information.

A fundamental assumption of conservation genetics is that census population size is correlated with N_e and that populations with small N_e experience elevated levels of genetic drift and inbreeding, which can threaten their survival (64). One consequence of small N_e is inbreeding depression, which refers to decreased fitness that occurs in small populations due to homozygosity of recessive deleterious alleles. Another is that the efficacy of negative selection is reduced, which can lead to an increase in frequency of deleterious mutations, increasing genetic load. Finally, reduced N_e is also correlated with reduced ability to adapt due to decreased additive genetic variance, which limits response to selection. An important principle of conservation genetics is therefore that population declines lead to reductions in N_e, which reduce population viability, and that measures to restore genetic variation can mitigate these effects (63).

The utility of neutral variation in conservation genetics is, however, subject to debate (63, 65, 66). Genetic variation correlates with conservation status as defined by the International Union for Conservation of Nature (IUCN) in eutherian mammals (67). However, in both animals and plants, this association is weak, and many endangered populations do not have low heterozygosity (68, 69). When controlling for phylogenetic relatedness, threatened animal and plant species typically have lower levels of heterozygosity than closely related nonthreatened species, a finding that supports the importance of genetic variation in population viability (70). However, levels of neutral genetic variation are not a consistent proxy for identifying endangered species (65). This is because many other factors apart from population size and viability govern heterozygosity, and levels of genetic diversity vary among phylogenetic groups.

In contrast to terrestrial vertebrates, insects commonly have large population sizes and densities, and many insects likely can maintain high N_e (71). For example, a study of a threatened grasshopper species revealed high heterozygosity despite a highly fragmented and disconnected distribution (72). This likely implies that insect species are at greater risk of being driven to extinction by a driver such as habitat loss before any negative genetic effects are experienced. Conservation genetic approaches for insects should be tailored for species that typically have high N_e. For example, it may be important to identify declining populations even before they experience negative genetic effects due to high levels of drift and inbreeding.

Comparison of levels of heterozygosity among animals has revealed that the overriding determinant of levels of genetic variation is intrinsic life-history traits rather than geographic range or population history (61, 69). The reproductive strategy of a species is the main factor that determines genetic diversity. Species with the highest levels of polymorphism tend to be r-strategists that rapidly produce large numbers of small eggs and do not invest in parental care. In contrast, K-strategists, which invest heavily in a small number of offspring, tend to have lower heterozygosity. A plausible, but untested, theory to explain these observations is that N_e depends most strongly on the minimum size of historical population fluctuations. This implies that K-strategists can recover from extreme population bottlenecks that would cause r-strategists to go extinct (61). Therefore, life history must be taken into account to interpret differences in N_e between species if one aims to uncover evidence for declines or identify at-risk species.

Insects display a wide range of life histories and ecological strategies that very likely affect levels of genetic variation (73). Social insects, such as ants, termites, and social bees and wasps, are K-strategists that make large investments in the colony and produce relatively small numbers of reproductively capable individuals. However, a larger proportion of insects, including pests like locusts, mosquitoes, and aphids, are rapidly expanding r-strategists. In addition to these major differences in reproductive strategies, interspecific variation in many other traits likely influences levels of heterozygosity. Additional reproductive traits vary from different forms of parthenogenesis to obligate sexual reproduction and in the number of life cycles per year. Both diploid and haplo-diploid sex-determination systems are in evidence. Insects also vary in their ability to

disperse on land, in water, and in the air. Insects occupy a wide range of ecological niches and positions in food webs and various forms of parasitic and nonparasitic lifestyles, with broad or narrow host specialization. Insects are also specialized in terms of food sources, with some pollinators being limited to specific plants and others being broad generalists.

So far, our understanding of how factors such as life histories and ecological niche breadth affect levels of genetic diversity in insect species is limited. Analysis of levels of genetic variation and genetic load in eusocial versus nonsocial insects found that levels of heterozygosity in four social species (two ants, one termite, and one bee) were similar to those found in vertebrates, whereas those in nonsocial insect species (two butterflies and a mosquito) were approximately 10 times higher (74). Furthermore, social insects display an excess of nonsynonymous polymorphism, indicating relaxed purifying selection. Interestingly, however, a study of purifying selection in 169 Hymenoptera species found that relaxation of selection was a feature of all bee species but not of other solitary Hymenoptera, which might suggest that a pollen-collecting lifestyle leads to low N_e (75). Life-history factors therefore hugely impact levels of genetic diversity among insects, although the underlying mechanisms are poorly understood.

The relationship between population declines, degree of vulnerability, and neutral genetic diversity is therefore complex. One way to improve our ability to diagnose conservation status of a population from levels of neutral genetic variation is to gain a better understanding of how intrinsic factors related to ecological strategies and life histories and extrinsic factors related to range and demographic history determine N_e in insects (69). This can be used to make baseline predictions of genetic variation expected for species with different life-history traits to identify those that deviate. Such expectations allow identification of species or populations with unexpectedly low N_e that could be of conservation concern. Furthermore, using population genomics, we can directly estimate levels of inbreeding and genetic load experienced by a population, which is potentially a more powerful way to identify vulnerable populations than measures of neutral genetic variation (2).

The intrinsic features of insects—high N_e, rapid reproduction, small body size, and high population densities—could mean that assays of neutral genetic variation lack power to identify insect populations of conservation concern. In addition, an important goal is to assess population trends in all species, not only those with drastically reduced N_e. Population genomics offers an array of techniques that move away from reliance on basic measures of neutral genetic variation, which could have great utility in measuring insect declines and identifying vulnerable populations.

4. POPULATION GENOMIC METHODS FOR INSECT CONSERVATION

Several specific goals are important for insect conservation. First, insect population trends are still understood poorly compared to those of vertebrates. It is imperative to accurately and comprehensively assess insect population trends by monitoring changes and inferring past population trends across species and regions. Second, we must identify species and populations threatened by high levels of genetic load, which can be investigated by quantifying levels of deleterious variation in populations. Third, we must identify cryptic biodiversity, consisting of previously unknown species. Population genomics is an essential tool for identifying new and cryptic species and identifying evolutionarily significant units (ESUs). Fourth, we need a better understanding of the drivers of insect declines and how insect populations are adapting to them to make accurate predictions of viability and future distributions, which is possible by incorporating genome scans for adaptive loci with models to predict changes in species distributions (76).

In this section, we review how population genomics can address these goals. Genomic methods can address questions previously addressed with a limited number of genetic markers with greater accuracy and precision, in some cases leading to different conclusions. These include estimating levels of genetic variation, detecting population fragmentation, and estimating population

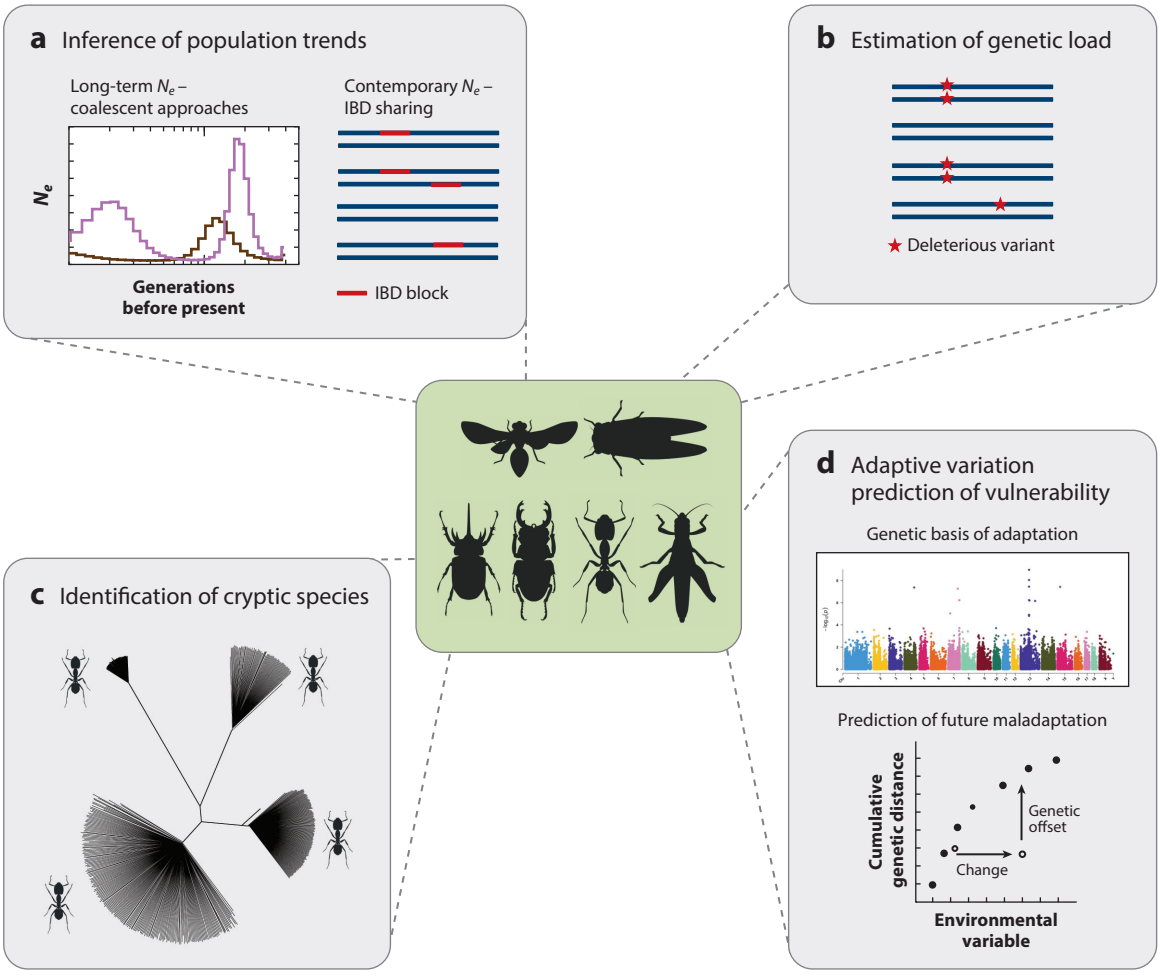


Figure 2

Four ways in which population genomic analyses can contribute to insect conservation. (*a*) Whole-genome variation data can be used to infer N_e in both long-term and contemporary timescales, which can uncover evidence for declines and identify threatened populations. (*b*) The distribution of deleterious mutations can be estimated, which can determine if populations have elevated detrimental genetic load. (*c*) Population-scale sequencing can delineate species and identify cryptic species. (*d*) Genome scans can identify genetic variants underlying adaptation to environmental conditions and stressors. This can be used to predict maladaptation of populations under future environmental conditions. Abbreviations: N_e, effective population size; IBD, identity by descent.

history and migration (1). In addition, genomic methods facilitate a range of additional analyses that are not possible without genome-wide analyses that could be particularly beneficial for insect conservation. These include a detailed reconstruction of population size fluctuations, direct estimation of deleterious genetic variation, precise species delineation, identification of the genetic basis of adaptation to stressors, and prediction of evolution in response to factors that drive declines (**Figure 2**).

A key resource for analyzing genome-wide variation is a reference genome assembly of the species of interest, which also facilitates analysis of genome variation in closely related species (77). High-quality reference genome assemblies with high contiguity, up to complete chromosomes, can now be produced using long-read sequencing technologies. Reference genome sequences

are available for a much smaller proportion of insect species compared to terrestrial vertebrates (**Figure 1**). However, several large-scale initiatives aim to generate reference sequences for all eukaryotes on Earth, including the Earth BioGenome Project, Darwin Tree of Life, and European Reference Genome Atlas (78), which will open up population genomics for a much larger fraction of Earth's biodiversity. With the help of a reference assembly, whole-genome sequencing data can be generated to assay variation in a large population sample of hundreds of individuals using high-throughput short-read sequencing (for example, using Illumina short-read sequencing technology).

4.1. Estimation of N_e and Population Demography

Analysis of genetic diversity is an important way to estimate N_e and infer population size changes in insect populations. Data on population trends from time-series data require major long-term research efforts to collect (6, 7). In addition, insects can have huge natural episodic population swings and can be highly migratory (e.g., monarch butterflies), which complicates analyzing population trends by surveying numbers in nature. Insects can also have multiple life stages that differ in many aspects (e.g., waterborne larvae and flying adults), and which stages should be counted to measure population trends most relevant to species viability is unclear. Inferring N_e from neutral genetic variation is therefore a powerful way to increase our understanding of insect population trends.

Importantly, N_e of a population is not a fixed value but rather a temporal continuum (79). A wide range of methods have been used to estimate N_e, which can be divided broadly into those that estimate either long-term or contemporary N_e. Long-term N_e can be estimated only with genetic data. Given an estimate of mutation rate, population genetic summary statistics based on nucleotide diversity such as Watterson's θ (80) or average heterozygosity per base can be used to estimate N_e. This gives a measure of long-term N_e, which can be defined as the harmonic mean of N_e over the time since the most recent common ancestor of sequences in the sample ($\sim 2N_e$ generations for diploids). This value reflects demographic changes since the time to the most recent common ancestor of the sequences in the population. If the population has experienced extensive population bottlenecks, this will be reflected in reduced long-term N_e. However, because this estimate also reflects much older population history, the effect may be subtle. Recent changes in N_e therefore may not be detectable using this statistic, and other methods are needed to accurately detect recent population declines (79).

4.1.1. Estimating past fluctuations in N_e using contemporary samples. The entire temporal trajectory of N_e can be inferred from genome-wide data. One possibility is to construct explicit demographic models of population history incorporating shifts in N_e (81). However, such models are necessarily complex and can provide only a simplistic picture of past population dynamics. A range of new nonparametric methods are now available to model past fluctuations in N_e in much greater detail using densely spaced markers in the genome. The first of these methods to be introduced was the pairwise sequentially Markovian coalescent (82) based on the coalescent hidden-Markov model (coalescent-HMM). The method analyzes the distribution of heterozygous sites in the genome of a single diploid individual. Using an HMM approach, it identifies blocks with distinct time to the most recent common ancestor and then models fluctuations in N_e based on the distribution of coalescent times of chromosomal segments.

One limitation of the pairwise sequentially Markovian coalescent is the lack of resolution on a timescale more recent than a few thousand generations. However, an array of related methods are now able to access more recent events (83–86). For example, the multiple sequentially Markovian coalescent method, also based on the sequentially Markovian coalescent, uses high-coverage phased data from a few diploid genomes, which enables inference of changes up to a few hundred

generations before present (85). Another approach, SMC++ (86), can take hundreds of unphased diploid genomes as input and infers historical variation in N_e using both the coalescent-HMM and analysis of the site-frequency spectrum and can infer N_e across the majority of time since the coalescence of the sequences in the sample. However, none of these coalescent-based methods can accurately infer N_e more recently than ~200 generations ago.

An important way to assess contemporary N_e is to directly measure changes in allele frequencies due to drift by sampling the population at more than one time point (87). This method is applicable for highly endangered populations in which N_e is small (<1,000) and drift is therefore easy to observe. However, the method is intractable for populations with large N_e, because huge sample sizes would be required to prevent sampling error in measuring allele frequencies greatly exceeding changes due to drift. Because even threatened insect species can have high N_e (72), this method likely has limited applicability to many insect populations.

Population history in the near past (~5–200 generations) can also be inferred by analyzing patterns of LD and the distribution of chromosomal segments of identity by descent (IBD) or identity by state. LD between pairs of single-nucleotide polymorphisms (SNPs) is always expected to decay with genetic distance, but the average level of LD between SNPs at different distances depends on N_e at different time points in the recent past. Several new approaches, such as LinkNe (88), SNeP (89), and GONE (90), extend traditional methods for estimating contemporary N_e using LD (91). Distribution of blocks of IBD in the genome also reflects recent N_e. Many long blocks of IBD shared between individuals indicate recent inbreeding or a low N_e in recent time, whereas many short IBD blocks indicate large contemporary N_e. Methods such as IBDNe analyze the distribution of IBD block lengths to infer N_e from a few to a few hundred generations in the past (92–94). In addition, methods based on analyzing the distribution of SNP frequencies are also powerful to infer recent demography (2, 81). These include δaδi, which fits demographic models to a diffusion approximation of the site-frequency spectrum (95).

The availability of genome-wide data from multiple individuals permits analysis of past fluctuations in N_e from a few generations ago until thousands of generations ago (96). Hence, analysis of genome-wide variation in contemporary populations could be a more efficient way to infer population trends than time-series collections for large numbers of insect species. These methods to infer variation in N_e through time to the present could potentially address some important issues for insect conservation: (*a*) The ability to infer recent changes in N_e using genetic data allows us to address insect declines without requiring long-term observational data. (*b*) We could determine the effects of major climatic events, for example, past ice ages, on N_e. (*c*) We could understand the influence of stressors on N_e by comparing timing of the introduction of these stressors with population fluctuations. (*d*) We could refine our understanding of how life-history traits influence N_e and demographic history (61, 69).

Examples of the utility of these approaches include an analysis of genome variation in the honeybee *Apis mellifera*, which indicated past declines in European populations during expanded ice sheets in Europe, when populations were confined to refugia, whereas the opposite trend was seen in African populations on the same timescale (97). Similar approaches have also been used to show that prehistoric climate change impacted populations of the oriental fruit moth *Grapholita molesta* (98). This provides a historical record of the effects of habitat loss. Population genomics was also used to track introductions and expansions of the invasive aquatic insect *Trichocorixa verticalis* in the western Mediterranean (99). Genomic inference was used to accurately predict the timing of a severe bottleneck in the endangered butterfly *Euphydryas gillettii* in North America, supporting the utility of genomic methods to infer recent population declines (100). Demographic inference in the diamondback moth, *Plutella xylostella*, indicated a decline since the last glacial maximum, followed by an expansion associated with global trade and its transformation into an agricultural

pest (101). Finally, genomic analysis of the agricultural pest *Bemisia tabaci* has revealed a complex demographic history and introgression between species involved in invasions (102).

4.1.2. Museum genomics. Genomics has also facilitated the ability to assess temporal changes in genetic diversity via sequencing of historical samples. Current high-throughput sequencing can recover sequences even when samples are highly degraded. In addition, museums across the world hold billions of biological samples, of which a large fraction are insects (103), and methods exist to extract DNA without damaging the specimens (104).

In some cases, these methods can be used to identify historical samples. For example, museum genomics has revealed that the Xerces blue butterfly (*Glaucopsyche xerces*) was a distinct species driven to extinction in North America (105). Another potentially important use of museum genomics is the ability to determine temporal changes in diversity (106). For highly endangered and fragmented species, it may be possible to detect loss of heterozygosity or increased fixation index due to extremely elevated genetic drift. For example, two endangered butterfly species with a fragmented distribution in Finland were shown via sequencing of museum specimens to have lost heterozygosity during the twentieth century (107).

However, for populations with high N_e, recent declines will likely not have a significant impact on overall levels of genetic variation compared to museum samples. It is therefore not straightforward to use museum samples to detect declines in insect species that are not severely endangered. For example, comparing genetic diversity in contemporary honeybees (*A. mellifera*) in Switzerland to those in museum samples revealed no substantial differences in Watterson's θ (108). However, European honeybees have long-term N_e of ~200,000 (97); therefore, population declines would likely not have a significant impact on θ unless they entailed a severe bottleneck. Analysis of temporal samples from museum specimens is most informative for the insect species that are most at risk from high levels of genetic drift.

4.2. Assessing Genetic Load by Identifying Deleterious Genetic Variants

All populations contain deleterious mutations, which persist for a period of time that depends on the intensity of drift and purifying selection. Population genetic theory indicates that strongly deleterious variants tend to be eliminated quickly and are unlikely to be present at high frequencies, whereas weakly deleterious variants may reach high frequencies and make a substantial contribution to the genetic load of a population (109, 110). Another factor that determines genetic load is the level of inbreeding, because it affects the number of recessive deleterious variants found as homozygotes, where they will be expressed and negatively affect fitness.

In general, genetic load is expected to be elevated in small and fragmented populations due to processes such as relaxation of selective constraint and increased levels of inbreeding. However, simulations show that demographic scenarios such as population bottlenecks, serial founder effects, and population expansions can have complex effects on the allele frequency spectrum and genetic load present in a population (110). Genetic drift may also be extreme at the leading edge of range expansions, leading to fixation of deleterious mutations (111). Another important effect is purging, the selective removal of deleterious variation that occurs in small populations due to selection against homozygotes (65). The existence of these effects means that genetic load cannot be predicted simply by assaying neutral variation. However, with population genomic data it is possible to directly identify potential deleterious mutations in individual genomes and estimate their effects on genetic load.

Estimating genetic load requires a well-annotated reference genome assembly and genome-wide variation data from the population of interest. With the sequence of an outgroup, we can determine which of the alleles at each SNP is ancestral. Derived alleles at functional sites are

assumed to be deleterious, but the severity varies and can be estimated computationally. Various tools exist to predict the effects of deleterious variants. For example, snpEff (112) categorizes mutations according to genome annotation and identifies nonsynonymous and loss-of-function variants. With additional related species, conservation scores such as GERP (Genomic Evolutionary Rate Profiling) scores (113) can be estimated across the genome, and the fitness effect of a SNP can be estimated using a mutation scoring method, such as the SIFT (Sorting Intolerant from Tolerant) algorithm (114). Additional methods that predict effects on protein structure and function are available but are applicable only to human proteins (115).

Using these algorithms, we can therefore estimate the number of deleterious mutations present in a genome and whether they are present as homo- or heterozygotes and use this to estimate a mutational burden. However, how to directly estimate these measures' effect on fitness remains unclear. An important factor in determining the effect of deleterious variation on fitness is the model of dominance. Most mutations known to cause disease in humans and animal models are recessive, which indicates that most deleterious mutations are recessive/loss of function. The distribution of dominance effects among deleterious mutations requires further investigation and is important to understand to predict reduction in fitness from deleterious mutations in the genome (63). To evaluate whether genetic load is elevated, it is usually necessary to compare the burden of deleterious mutations with a baseline. This could be a closely related species that has not suffered a population decline, or alternatively a historical pre-decline population represented by museum samples (106).

In addition to increasing genetic load, population bottlenecks can also lead to purging, because they cause deleterious recessive mutations to occur in homozygotes, which both increases genetic load and can lead to their removal from the population. Purging can be a positive effect of low N_e observed in bottlenecked mammals, such as the Iberian lynx (116), and a meta-analysis of threatened species suggests it could be a common process that ameliorates the effects of deleterious variation (117). However, its importance in the viability of insect populations is unclear. More studies are needed to understand the prevalence of purging. In insects with a haploid stage, such as Hymenoptera, where males are haploid, recessive mutations will always be unmasked, and those with severe effects will be strongly selected against even in the absence of inbreeding (118). However, haplodiploids suffer the effects of inbreeding via diploid male production due to homozygosity at the sex-determining locus (119).

The methods presented here are extremely promising for application to insect conservation and are a more direct way of assaying genetic load than using neutral genetic variation. However, directly predicting how these measures relate to fitness remains challenging, and functional annotations and conservation scores required to determine the effects of mutations are not available for most insect genomes. More data are needed from multiple species to develop predictions of genetic load among taxonomic groups.

The significance of high genetic load may depend on multiple factors, such as mating strategy and dispersal ability. For example, a small population of the butterfly *Melitaea cinxia* isolated on a small island exhibited high genetic load in fitness-related traits, implying extensive fixation of deleterious recessive mutations (120). However, some insect species, such as the fire ant *Solenopsis invicta* (121) and solitary bee *Lasioglossum leucozonium* (122), have been involved in successful biological invasions despite suffering extreme genetic load and high diploid male production. The connection between genetic load and population vulnerability is therefore not always straightforward.

4.3. Defining Evolutionarily Significant Units for Conservation

Species delineation is fundamental for conservation, and lists of endangered species (e.g., in the form of Red Lists) are central tools for protecting biodiversity. Genetic research can clearly

contribute to this through phylogenetic analysis to delineate taxa. In addition, the Barcode of Life project aims to generate DNA barcodes for vouchered specimens, which is a powerful way to catalog insect species diversity (123). However, despite the power and efficacy of these methods, there is no one-to-one mapping between molecular barcode and species identity. Given the process of incomplete lineage sorting, gene trees from a single locus often do not correspond to species relationships and may lack information to distinguish species (124). Population genomics is therefore a more precise way to delineate species.

One challenge is the potentially large number of cryptic insect species (125). These species may go unnoticed by taxonomists but are revealed by the presence of unexpected genetic clusters. Hidden cryptic species can be identified by sequencing large population samples. For example, a study using DNA barcoding identified 10 previously hidden cryptic species in the neotropical butterfly *Astraptes fulgerator* (126). Whole-genome sequencing of 281 bumblebee samples identified originally as *Bombus sylvicola* from Colorado identified a distinct genetic cluster that corresponded to no other known species and was not detected by previous genetic studies (127, 128). Individuals from this cluster live in sympatry with *B. sylvicola*, but no evidence of recent hybridization was identified, indicating the presence of a previously unidentified species. Further large-scale population genomic surveys in insects are certain to identify more examples of hidden cryptic species, which represent important units of conservation.

Another difficulty with delineating species is that in nature, individuals do not cluster into distinct entities, and no universally agreed criteria exist to define species. This is further complicated by findings from speciation genomics indicating that species boundaries are fluid (129). Population genomics shows that genomes of closely related species are mosaics of different ancestries, and many examples now indicate that hybridization and gene flow across species boundaries are common. Prolific exchange of genes occurs between species in the rapidly expanding *Heliconius* neotropical butterfly genus, particularly at genes involved in coloration and mimicry (130). *Anopheles* mosquitoes show different levels and patterns of relatedness across the genome, with no consistent species delineation (131). These studies argue against species as distinct entities and suggest a view of speciation as a continuum. Genomics could assist species delineation by establishing norms based on common criteria from reference systems (132). This would prevent conservation efforts from unduly prioritizing well-studied and oversplit taxa.

Nature consists of evolving populations rather than immutable species. Despite its clear utility, the inherent ambiguity of species in nature suggests that species is insufficient as the only unit of conservation (133). Additional criteria could be used to define ESUs of conservation. For instance, genetic distinctiveness could be used as criteria for conservation value (134). An advantage of this approach is that it would assign conservation value to highly isolated, but distinct, populations of species that have a wider distribution elsewhere. Such examples are *Erebia pandrose*, a butterfly with a tiny, isolated population that is genetically distinct in the Apennines (135); the butterfly *Parnassius apollo*, with numerous, highly localized subspecies in Europe (136); or the flightless halobiontic groundbeetles of the genus *Pogonus*, with highly isolated populations in extreme niche habitats in central Europe (137).

4.4. Genetic Adaptation and Evolutionary Responses to Drivers of Declines

Insect populations can respond to drivers of decline such as habitat loss, pesticides, and climate change in several ways. First, if they are sufficiently mobile, they can move to track optimal conditions, as has been observed in range shifts in various insect species such as bumblebees (47) and butterflies (138). Genetic tools can be useful for measuring this response, as dispersal leads to gene flow (139). Second, individuals can persist via plastic responses constrained by their physiological limits, which can be investigated through acclimation and life-history experiments using

populations from different environments. Third, populations may genetically adapt to changing conditions, provided sufficient heritable variation in fitness-related traits is present (140).

An understanding of the genetic basis of adaptation is needed to predict how populations will evolve to stressors. Several methods exist to identify the genetic basis of adaptation. If the trait of interest that influences fitness is known, then quantitative trait loci mapping or genome-wide association studies can be used to map the genetic basis of the trait. Other methods can be used if the phenotypic basis for adaptation is not known, or even if it is unclear a priori that populations are locally adapted. This involves statistical comparison of genetic variation across the genomes of populations that reside in different environments, which can be either discrete locations or situated along a cline (e.g., latitudinal or altitudinal). Correlations with environmental variables can also be identified via gene-environment association analysis (GEA) (140, 141).

GEA aims to identify associations between allele frequencies and environmental variables that indicate local adaptation to the environment. Fst outlier approaches have been a common tool, but they do not adequately control for false positives due to population structure (142). This can be done via methods such as BAYENV2 (143, 144), which uses mixed-effect models in a Bayesian framework and tests whether models including environmental factors explain data significantly better than those containing only neutral genetic structure. Another method uses latent factor mixed models, in which neutral genetic variation is incorporated as a latent factor using the program LFMM (145). Another possibility is redundancy analysis, which models multiple environmental predictors simultaneously in a multivariate approach (146).

4.4.1. Climate change. Understanding adaptation along climatic gradients is important to predict the effect of climate change. A wealth of studies in natural populations of the fruit fly *Drosophila* have been instrumental in our understanding of climate adaptation in insects. These have revealed parallel variation at specific loci, including chromosomal inversions, along clines on different continents, in some cases related to known traits (147, 148). In addition, adaptive oscillations in allele frequencies at specific SNPs occur through seasons of the year (149). This exemplifies how short generation times in insects allow them to adapt rapidly to environmental change. Shifts in allele frequency also have been linked to climate change (150).

In the western honeybee *A. mellifera*, adaptation to climate appears to have mainly a polygenic basis. This is inferred by analyzing adaptation along latitudinal clines in honeybees of mixed ancestry in the Americas, which demonstrates parallel variation in allele frequencies across the whole genome (151). However, in another example, two megabase-scale chromosomal inversions are found at high frequencies in honeybees at high elevations in East Africa, where they govern environmental adaptation (152, 153). In the eastern honeybee *Apis cerana*, adaptation to high-altitude environments appears to have a polygenic basis (154).

In bumblebees, analysis of heat and drought tolerance, using comparative genomics, transcriptomics, and experimental assays, identified genes and pathways involved in climate adaptation in samples from populations living in different climates (155). In the cosmopolitan diamondback moth, climate-associated adaptive variation shows a latitudinal pattern, and a key temperature-responsive gene was identified through gene editing (156). Seven stonefly species in Japan showed local adaptation across a latitudinal gradient, revealing more than 290 associated variants from a reduced representation genomics scan (ddRAD) (157). GEA was used to identify genes with significant correlations to climatic variables in the damselfly *Ischnura elegans*, including heat-shock proteins (158).

4.4.2. Pesticides. Research into the genetic basis of pesticide resistance has focused mainly on pests and disease vectors but will likely also be informative regarding other insects. Through

population genomics, an array of genes have been identified that govern resistance to pesticides in *Anopheles* mosquitoes (159). In this case, this information can be used to understand the effects of pesticide treatment and design effective pest control strategies. For neonicotinoids, there is evidence that agricultural pests are developing resistance (160), but the effect selection pressure by these pesticides is having on most (nontarget) insects is unclear. Sublethal effects of pesticides clearly are of great importance (161).

In the Colorado potato beetle, *Leptinotarsa decemlineata*, genome resequencing showed repeated selection on insecticide resistance from standing variation, involving similar pathways but different genes (162). In the psyllid *Bactericera cockerelli*, insecticide resistance was associated with more than 400 loci (163). A full genome scan in the aphid *Myzus persicae* revealed a high diversity of resistance mutations segregating in global populations (164). Genomic analyses have shown that the invasive moth *Helicoverpa armigera* transferred an insecticide-resistance allele into the local *Helicoverpa zea* via adaptive introgression (165). These studies reveal important mechanisms of how wild populations could respond to the presence of pesticides.

4.4.3. Other drivers. Habitat loss and degradation are key drivers of insect losses, and population genomics can be used to understand their effects. These can involve adaptation to new environmental conditions or hosts. Transcriptomic analysis has revealed candidate genes for adaptation to a novel host plant and drier habitats in a rare cicada (166), and adaptation to host plants in some herbivorous hemipterans has been showed to be controlled genetically (167). Genome scans have identified the genetic basis of insect traits involved in host specialization in specialist versus generalist moths (168) and quantitative trait loci controlling feeding of pea aphids on different host plants (169). Furthermore, genome scans revealed parallel changes in pairs of populations of stick insects adapted to preferred and alternative host plants, which also drive reproductive isolation (170). In addition to identifying adaptation, genetic markers can be used to determine relatedness and trace lineage survival in different habitats. In bumblebees, these methods have been used to show that lineage survival is enhanced in habitats with rich foraging (171).

The genomic basis of adaptation to pathogens has been studied intensively in honeybees (*A. mellifera*) in relation to traits that mediate resistance or tolerance to the *Varroa destructor* mite (172). Specific traits identified in honeybees, including removal of infected brood by worker bees, have likely been under selection since the global spread of *V. destructor* starting in the 1950s (173), and the genetic basis of some of these traits is beginning to be understood (174). However, parasite resistance in managed honeybee colonies likely occurs through different mechanisms than in wild solitary bees; therefore, we must understand more about adaptation to pathogens in other insects (175). Immune genes are particularly fast evolving, and expansion and losses are believed to be important ways for insects to adapt to new pathogens (176).

4.5. Incorporating Evolution into Species Conservation

Information about the genetic basis of adaptation can be used for conservation in several ways. Firstly, sets of adaptive genetic variants can have intrinsic conservation value (177). Examples include chromosomal inversions that define ecotypes with adaptation to local environments and different migration patterns (178). Such variants could be considered to be analogous to ESUs. The presence of such functional variation influences species' capacity to adapt to environmental change, which is important for evolutionary rescue and prediction of selection response. In addition, genomic studies of adaptation provide information about molecular pathways that are important in response to stressors.

Another key use for this information lies in incorporating local adaptation into species distribution models that seek to predict distributions of species in future environmental conditions.

The concept of genomic vulnerability, or genetic offset (76, 140, 179), is that by using the statistical relationship between frequency of adaptive alleles and local environmental conditions along a transect, one can estimate the optimal set of allele frequencies in a hypothetical future environment. Genetic offset is the distance between the current and required genetic composition of a set of adaptive loci under predicted future environmental conditions. A large genetic offset under a certain future scenario implies that the population is at high risk of maladaptation and is considered vulnerable.

The most prominent examples of using genomics to predict evolutionary responses to climate change come from outside insects. For example, analysis of genomic variation in a species of coral showed that the degree of bleaching expected due to warming oceans depends on local genotypes at specific loci (180). In the plant *Arabidopsis thaliana*, drought response depends strongly on how individuals are genetically adapted to local conditions (181). In yellow warblers, variation at certain genes is associated with local adaptation across its breeding range. Strikingly, populations that were predicted to require the most evolution of allele frequency change to keep pace with future climate change were also those found to already be declining (182). These findings strongly suggest that integrating genomic adaptation measures into predictive models will be vital for accurate inference of climate change responses.

Species distribution models are an important way to predict future species distributions under future projections of climate change. For example, in European bumblebees, an atlas of climatic risk has been produced (49). However, such predictions consider neither local adaptation nor the potential for evolution. If populations show extreme local adaptation across their range, then climate change will lead to loss of fitness throughout the range. This is particularly problematic for flightless and relatively immobile species, which may be unable to track their environmental niches to keep pace with climate change. For highly mobile species, genomic offset methods can be used to predict optimal genotypes under future climate scenarios, which can then be validated by genetic monitoring of gene flow as the climate changes.

5. FUTURE PROSPECTS AND CHALLENGES FOR INTEGRATING GENOMICS TO FIGHT INSECT DECLINES

The unprecedented loss of biodiversity in the Anthropocene is arguably comparable to the mass extinction that wiped out dinosaurs 65.5 Mya (11). It is estimated that approximately half of the species on earth are insects, but loss of biodiversity in insects is far less understood than in larger animals and plants (6). However, the available data suggest that insect declines could be even more extreme than those of other taxonomic groups. Population genomics offers a powerful new tool for insect conservation (2, 140), enabled by the huge numbers of reference genome assemblies that will soon be available.

In this review, we have outlined issues in which population genomics can contribute to insect conservation (**Figure 1**). The first is the accurate measurement of population declines. Directly assaying temporal changes in population sizes in the vast number of insect species by collecting time-series data is impossible. However, by assaying genomic variation in present-day populations, we can infer population size fluctuations even in the recent past (79, 81). This provides evidence for both recent population declines and ancient changes due to climatic fluctuations. By integrating results from these analyses in a diverse range of species across insect orders, we could address some key questions. For example, do common trends in population history across species correlate with prehistoric climate change? Do different insect orders and geographical regions show similar evidence for declines in the Anthropocene? How is N_e affected by factors such as species-intrinsic traits, habitat, and range? These analyses will aid in identifying specific at-risk species and allow

the development of a metric to predict population vulnerability based on expected levels of genetic variation in relation to life-history traits.

A second application of population genomics is the ability to estimate genetic load. This is potentially a more reliable way to identify populations under threat than estimating N_e from neutral variation, which is commonly done via classical conservation genetics (2, 65, 106). However, no standardized way to measure genetic load or determine what level is hazardous to a population exists. Therefore, these measures must be generated in a large range of species to determine critical values. This will require accurate annotation of genome assemblies and estimation of base conservation scores across insect genomes, similar to those available for mammals (67).

A third application of population genomics is the increased resolution in delineation of species and identification of cryptic species. This is particularly useful in taxonomic groups of insects that are difficult to distinguish morphologically. A wide range of examples from speciation genomics indicate that genomes of closely related species are mosaics of ancestry from incomplete lineage sorting and hybridization (132). Population genomics studies will therefore aid in species delineation and ESU identification.

The fourth application is identifying the genetic basis of adaptation. Determining how populations are adapted to their environments can guide monitoring and management decisions. For example, by defining specific alleles or blocks of coadapted alleles (for example, inversions) involved in certain adaptations, we can understand a population's capacity to adapt to change and act to conserve specific adaptations. In addition, genomic offset methods (179) allow incorporation of evolution into models of species distribution, by predicting which individuals will be tolerant to certain environments. We are unaware of any examples of the application of these methods in insects, but they have the potential to improve our ability to predict future species distributions and identify populations vulnerable to the effects of climate change.

Scaling up population genomic approaches to study the millions of insect species presents major challenges. Currently, entire arthropod communities can be surveyed using metabarcoding, which entails short-read sequencing of bulk samples from malaise traps (183). Bulk sequencing can also be applied to eDNA (environmentally sampled DNA). Further developments in sequencing technology and the availability of reference genomes from a large fraction of eukaryotic life, and improved algorithms for bioinformatics and population genetic inference, should enable population genomic analyses using whole-genome sequencing of bulk samples. Another important advance is the use of artificial intelligence in image analyses, which allows automatic species identification from malaise traps (32) and could be coupled with sequence analysis to link genotype and phenotype.

Genetic monitoring of insect populations could also be an important tool for their conservation. Such programs could be particularly informative when used to monitor changes in adaptive allele frequency. For example, genomic offset methods can be used to predict the optimal allele frequencies at adaptive loci under certain environmental conditions, and changes at these loci can be monitored over time. Genetic monitoring can also be used to track changes at deleterious alleles and to manage the presence of specific adaptive variants, such as those involved in insecticide resistance.

Finally, genomic technologies could be used to directly manipulate wild insect populations. For example, gene drive technology based on CRISPR-CAS9 gene editing could introduce adaptive alleles to allow wild populations to adapt to stressors, such as introducing pesticide resistance or alleles that facilitate adaptation to climate change. They could also be used to introduce deleterious alleles in pests. Furthermore, cloning technology could potentially be used in future to resurrect extinct species (184). However, we must proceed with extreme caution to avoid unwanted effects.

Genomics is finding many new applications in conservation, but studies in insects are still lagging behind those in vertebrates. Although reports of insect declines are alarming, the full details and underlying causes are still unclear. Population genomics can aid our understanding of insect population trends and their drivers and inform the design of conservation programs. In conclusion, genomics offers an extremely powerful tool kit to tackle insect declines, and continued rapid progress is crucial to address this urgent issue.

SUMMARY POINTS

1. Several studies have identified drastic losses in insect numbers and biodiversity. Globally insect declines may be even more extensive than seen in better-surveyed groups of animals and plants.
2. Habitat loss, pesticides, and climate change are likely the most important drivers of insect declines.
3. Population genomics offers many tools to investigate insect declines and predict how populations will adapt to future threats, but very few studies are available so far.
4. Levels of neutral genetic diversity are more strongly impacted by intrinsic life-history traits than species conservation status. A better understanding of the determinants of levels of genetic variation is needed to use measures of genetic variation to predict population vulnerability.
5. Conservation genomic analyses should be tailored to intrinsic features of insects such as short generation times, rapid reproduction, and large populations.
6. Analysis of whole-genome variation can be used to infer historical population trends and estimate levels of genetic load to identify threatened species.
7. Analysis of intraspecific genome variation can identify previously hidden cryptic species or locally adapted subpopulations of conservation value.
8. Identification of the genetic basis of local adaptation allows prediction of the effects of drivers such as climate change on insect populations, taking evolution in account.

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LITERATURE CITED

1. Supple MA, Shapiro B. 2018. Conservation of biodiversity in the genomics era. *Genome Biol.* 19:131
2. Hohenlohe PA, Funk WC, Rajora OP. 2021. Population genomics for wildlife conservation and management. *Mol. Ecol.* 30(1):62–82
3. Primmer CR. 2009. From conservation genetics to conservation genomics. *Ann. N.Y. Acad. Sci.* 1162:357–68

4. Allendorf FW, Hohenlohe PA, Luikart G. 2010. Genomics and the future of conservation genetics. *Nat. Rev. Genet.* 11(10):697–709
5. Ouborg NJ, Pertoldi C, Loeschcke V, Bijlsma RK, Hedrick PW. 2010. Conservation genetics in transition to conservation genomics. *Trends Genet.* 26(4):177–87
6. **Wagner DL. 2020. Insect declines in the Anthropocene. *Annu. Rev. Entomol.* 65:457–80**
7. Sánchez-Bayo F, Wyckhuys KAG. 2019. Worldwide decline of the entomofauna: a review of its drivers. *Biol. Conserv.* 232:8–27
8. Hallmann CA, Sorg M, Jongejans E, Siepel H, Hofland N, et al. 2017. More than 75 percent decline over 27 years in total flying insect biomass in protected areas. *PLOS ONE* 12(10):e0185809
9. Seibold S, Gossner MM, Simons NK, Blüthgen N, Müller J, et al. 2019. Arthropod decline in grasslands and forests is associated with landscape-level drivers. *Nature* 574(7780):671–74
10. van Klink R, Bowler DE, Gongalsky KB, Swengel AB, Gentile A, Chase JM. 2020. Meta-analysis reveals declines in terrestrial but increases in freshwater insect abundances. *Science* 368(6489):417–20
11. Dirzo R, Young HS, Galetti M, Ceballos G, Isaac NJB, Collen B. 2014. Defaunation in the Anthropocene. *Science* 345(6195):401–6
12. IPBES. 2019. *Global Assessment Report on Biodiversity and Ecosystem Services of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services*, ed. ES Brondizio, J Settele, S Díaz, HT Ngo. Bonn, Ger.: IPBES Secr.
13. Scheffers BR, Joppa LN, Pimm SL, Laurance WF. 2012. What we know and don't know about Earth's missing biodiversity. *Trends Ecol. Evol.* 27(9):501–10
14. Stork NE. 2018. How many species of insects and other terrestrial arthropods are there on earth? *Annu. Rev. Entomol.* 63:31–45
15. Bowler DE, Heldbjerg H, Fox AD, de Jong M, Böhning-Gaese K. 2019. Long-term declines of European insectivorous bird populations and potential causes. *Conserv. Biol.* 33(5):1120–30
16. Lister BC, Garcia A. 2018. Climate-driven declines in arthropod abundance restructure a rainforest food web. *PNAS* 115(44):E10397–406
17. Powney GD, Carvell C, Edwards M, Morris RKA, Roy HE, et al. 2019. Widespread losses of pollinating insects in Britain. *Nat. Commun.* 10:1018
18. Goulson D, Nicholls E, Botías C, Rotheray EL. 2015. Bee declines driven by combined stress from parasites, pesticides, and lack of flowers. *Science* 347(6229):1255957
19. Potts SG, Biesmeijer JC, Kremen C, Neumann P, Schweiger O, Kunin WE. 2010. Global pollinator declines: trends, impacts and drivers. *Trends Ecol. Evol.* 25:345–53
20. Biesmeijer JC, Roberts SPM, Reemer M, Ohlemüller R, Edwards M, et al. 2006. Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science* 313(5785):351–54
21. Thomas JA, Telfer MG, Roy DB, Preston CD, Greenwood JJD, et al. 2004. Comparative losses of British butterflies, birds, and plants and the global extinction crisis. *Science* 303(5665):1879–81
22. Franzén M, Johannesson M. 2007. Predicting extinction risk of butterflies and moths (Macrolepidoptera) from distribution patterns and species characteristics. *J. Insect Conserv.* 11(4):367–90
23. Conrad KF, Warren MS, Fox R, Parsons MS, Woiwod IP. 2006. Rapid declines of common, widespread British moths provide evidence of an insect biodiversity crisis. *Biol. Conserv.* 132(3):279–91
24. Neumann P, Carreck NL. 2010. Honey bee colony losses. *J. Apic. Res.* 49(1):1–6
25. Kerr JT, Pindar A, Galpern P, Packer L, Potts SG, et al. 2015. Climate change impacts on bumblebees converge across continents. *Science* 349(6244):177–80
26. Bartomeus I, Ascher JS, Gibbs J, Danforth BN, Wagner DL, et al. 2013. Historical changes in northeastern US bee pollinators related to shared ecological traits. *PNAS* 110(12):4656–60
27. Wagner DL. 2019. Global insect decline: comments on Sánchez-Bayo and Wyckhuys 2019. *Biol. Conserv.* 233:332–33
28. Thomas CD, Jones TH, Hartley SE. 2019. "Insectageddon": a call for more robust data and rigorous analyses. *Glob. Change Biol.* 25(6):1891–92
29. Pilotto F, Kühn I, Adrian R, Alber R, Alignier A, et al. 2020. Meta-analysis of multidecadal biodiversity trends in Europe. *Nat. Commun.* 11:3486
30. Crossley MS, Meier AR, Baldwin EM, Berry LL, Crenshaw LC, et al. 2020. No net insect abundance and diversity declines across US Long Term Ecological Research sites. *Nat. Ecol. Evol.* 4(10):1368–76

6. Balanced and comprehensive summary of the evidence for insect declines and their drivers.

31. Welti EAR, Joern A, Ellison AM, Lightfoot DC, Record S, et al. 2021. Studies of insect temporal trends must account for the complex sampling histories inherent to many long-term monitoring efforts. *Nat. Ecol. Evol.* 5(5):589–91
32. Wagner DL, Grames EM, Forister ML, Berenbaum MR, Stopak D. 2021. Insect decline in the Anthropocene: death by a thousand cuts. *PNAS* 118(2):e2023989118
33. Outhwaite CL, McCann P, Newbold T. 2022. Agriculture and climate change are reshaping insect biodiversity worldwide. *Nature* 605(7908):97–102
34. Hansen MC, Potapov PV, Moore R, Hancher M, Turubanova SA, et al. 2013. High-resolution global maps of 21st-century forest cover change. *Science* 342(6160):850–53
35. Carrasco LR, Webb EL, Symes WS, Koh LP, Sodhi NS. 2017. Global economic trade-offs between wild nature and tropical agriculture. *PLOS Biol.* 15(7):e2001657
36. Zanon M, Davis BAS, Marquer L, Brewer S, Kaplan JO. 2018. European forest cover during the past 12,000 years: a palynological reconstruction based on modern analogs and remote sensing. *Front. Plant Sci.* 9:253
37. Kaplan JO, Krumhardt KM, Zimmermann N. 2009. The prehistoric and preindustrial deforestation of Europe. *Quat. Sci. Rev.* 28(27):3016–34
38. Kaplan JO, Krumhardt KM, Gaillard M-J, Sugita S, Trondman A-K, et al. 2017. Constraining the deforestation history of Europe: evaluation of historical land use scenarios with pollen-based land cover reconstructions. *Land* 6(4):91
39. Botías C, David A, Hill EM, Goulson D. 2017. Quantifying exposure of wild bumblebees to mixtures of agrochemicals in agricultural and urban landscapes. *Environ. Pollut.* 222:73–82
40. Ollerton J, Erenler H, Edwards M, Crockett R. 2014. Extinctions of aculeate pollinators in Britain and the role of large-scale agricultural changes. *Science* 346(6215):1360–62
41. Henry M, Béguin M, Requier F, Rollin O, Odoux J-F, et al. 2012. A common pesticide decreases foraging success and survival in honey bees. *Science* 336(6079):348–50
42. Gill RJ, Ramos-Rodriguez O, Raine NE. 2012. Combined pesticide exposure severely affects individual- and colony-level traits in bees. *Nature* 491(7422):105–8
43. Whitehorn PR, O'Connor S, Wackers FL, Goulson D. 2012. Neonicotinoid pesticide reduces bumble bee colony growth and queen production. *Science* 336(6079):351–52
44. Rundlöf M, Andersson GKS, Bommarco R, Fries I, Hederström V, et al. 2015. Seed coating with a neonicotinoid insecticide negatively affects wild bees. *Nature* 521(7550):77–80
45. Goulet H, Masner L. 2017. Impact of herbicides on the insect and spider diversity in eastern Canada. *Biodiversity* 18(2–3):50–57
46. Öckinger E, Hammarstedt O, Nilsson SG, Smith HG. 2006. The relationship between local extinctions of grassland butterflies and increased soil nitrogen levels. *Biol. Conserv.* 128(4):564–73
47. Soroye P, Newbold T, Kerr J. 2020. Climate change contributes to widespread declines among bumble bees across continents. *Science* 367(6478):685–88
48. Breed GA, Stichter S, Crone EE. 2013. Climate-driven changes in northeastern US butterfly communities. *Nat. Clim. Change* 3(2):142–45
49. Rasmont P, Franzén M, Lecocq T, Harpke A, Roberts SPM, et al. 2015. Climatic risk and distribution atlas of European bumblebees. *BioRisk* 10:1–236
50. Forister ML, Novotny V, Panorska AK, Baje L, Basset Y, et al. 2015. The global distribution of diet breadth in insect herbivores. *PNAS* 112(2):442–47
51. Gérard M, Vanderplanck M, Wood T, Michez D. 2020. Global warming and plant-pollinator mismatches. *Emerg. Top. Life Sci.* 4(1):77–86
52. Miller-Struttmann NE, Geib JC, Franklin JD, Kevan PG, Holdo RM, et al. 2015. Functional mismatch in a bumble bee pollination mutualism under climate change. *Science* 349(6255):1541–44
53. Cornelissen B, Neumann P, Schweiger O. 2019. Global warming promotes biological invasion of a honey bee pest. *Glob. Change Biol.* 25(11):3642–55
54. Salih AAM, Baraibar M, Mwangi KK, Artan G. 2020. Climate change and locust outbreak in East Africa. *Nat. Clim. Change* 10(7):584–85
55. Wagner DL, Van Driesche RG. 2010. Threats posed to rare or endangered insects by invasions of nonnative species. *Annu. Rev. Entomol.* 55:547–68

44. Provides evidence that wild bees are negatively impacted by treatment of nearby crops with neonicotinoid pesticides at realistic doses.

56. Bertelsmeier C, Ollier S, Liebhold A, Keller L. 2017. Recent human history governs global ant invasion dynamics. *Nat. Ecol. Evol.* 1:0184
57. Fürst MA, McMahon DP, Osborne JL, Paxton RJ, Brown MJF. 2014. Disease associations between honeybees and bumblebees as a threat to wild pollinators. *Nature* 506(7488):364–66
58. Proesmans W, Albrecht M, Gajda A, Neumann P, Paxton RJ, et al. 2021. Pathways for novel epidemiology: plant-pollinator-pathogen networks and global change. *Trends Ecol. Evol.* 36(7):623–36
59. Arbetman MP, Meeus I, Morales CL, Aizen MA, Smagghe G. 2013. Alien parasite hitchhikes to Patagonia on invasive bumblebee. *Biol. Invasions* 15(3):489–94
60. Vilcinskas A. 2019. Pathogens associated with invasive or introduced insects threaten the health and diversity of native species. *Curr. Opin. Insect Sci.* 33:43–48
61. Romiguier J, Gayral P, Ballenghien M, Bernard A, Cahais V, et al. 2014. Comparative population genomics in animals uncovers the determinants of genetic diversity. *Nature* 515(7526):261–63
62. Leffler EM, Bullaughey K, Matute DR, Meyer WK, Ségurel L, et al. 2012. Revisiting an old riddle: What determines genetic diversity levels within species? *PLOS Biol.* 10(9):e1001388
63. Willi Y, Kristensen TN, Sgrò CM, Weeks AR, Ørsted M, Hoffmann AA. 2022. Conservation genetics as a management tool: the five best-supported paradigms to assist the management of threatened species. *PNAS* 119(1):e2105076119
64. Husemann M, Zachos FE, Paxton RJ, Habel JC. 2016. Effective population size in ecology and evolution. *Heredity* 117(4):191–92
65. Teixeira JC, Huber CD. 2021. The inflated significance of neutral genetic diversity in conservation genetics. *PNAS* 118(10):e2014096118
66. Kardos M, Armstrong EE, Fitzpatrick SW, Hauser S, Hedrick PW, et al. 2021. The crucial role of genome-wide genetic variation in conservation. *PNAS* 118(48):e2104642118
67. Genereux DP, Serres A, Armstrong J, Johnson J, Marinescu VD, et al. 2020. A comparative genomics multitool for scientific discovery and conservation. *Nature* 587(7833):240–45
68. Perry GH, Melsted P, Marioni JC, Wang Y, Bainer R, et al. 2012. Comparative RNA sequencing reveals substantial genetic variation in endangered primates. *Genome Res.* 22(4):602–10
69. Ellegren H, Galtier N. 2016. Determinants of genetic diversity. *Nat. Rev. Genet.* 17(7):422–33
70. Spielman D, Brook BW, Frankham R. 2004. Most species are not driven to extinction before genetic factors impact them. *PNAS* 101(42):15261–64
71. Tscharntke T, Steffan-Dewenter I, Kruess A, Thies C. 2002. Characteristics of insect populations on habitat fragments: a mini review. *Ecol. Res.* 17(2):229–39
72. Hoffmann AA, White VL, Jasper M, Yagui H, Sinclair SJ, Kearney MR. 2021. An endangered flightless grasshopper with strong genetic structure maintains population genetic variation despite extensive habitat loss. *Ecol. Evol.* 11(10):5364–80
73. Resh VH, Cardé RT, eds. 2009. *Encyclopedia of Insects*. Amsterdam: Academic. 2nd ed.
74. Romiguier J, Lourenco J, Gayral P, Faivre N, Weinert LA, et al. 2014. Population genomics of eusocial insects: the costs of a vertebrate-like effective population size. *J. Evol. Biol.* 27(3):593–603
75. Weyna A, Romiguier J. 2021. Relaxation of purifying selection suggests low effective population size in eusocial Hymenoptera and solitary pollinating bees. *Peer Community J.* 1:e2
76. Capblancq T, Fitzpatrick MC, Bay RA, Exposito-Alonso M, Keller SR. 2020. Genomic prediction of (mal)adaptation across current and future climatic landscapes. *Annu. Rev. Ecol. Evol. Syst.* 51:245–69
77. Paez S, Kraus RHS, Shapiro B, Gilbert MTP, Jarvis ED, Vert. Genomes Proj. Consort. 2022. Reference genomes for conservation. *Science* 377(6604):364–66
78. Lewin HA, Richards S, Lieberman Aiden E, Allende ML, Archibald JM, et al. 2022. The Earth BioGenome Project 2020: starting the clock. *PNAS* 119(4):e2115635118
79. Nadachowska-Brzyska K, Konczal M, Babik W. 2022. Navigating the temporal continuum of effective population size. *Methods Ecol. Evol.* 13(1):22–41
80. Watterson GA. 1975. On the number of segregating sites in genetical models without recombination. *Theor. Popul. Biol.* 7(2):256–76
81. Bourgeois YXC, Warren BH. 2021. An overview of current population genomics methods for the analysis of whole-genome resequencing data in eukaryotes. *Mol. Ecol.* 30(23):6036–71

61. Evidence that levels of genetic diversity across mammals are determined mainly by intrinsic life-history traits.

79. Comprehensive review of methods to infer historical N_e over a range of timescales.

82. Li H, Durbin R. 2011. Inference of human population history from individual whole-genome sequences. *Nature* 475(7357):493–96

83. Palacios JA, Wakeley J, Ramachandran S. 2015. Bayesian nonparametric inference of population size changes from sequential genealogies. *Genetics* 201(1):281–304
84. Speidel L, Forest M, Shi S, Myers SR. 2019. A method for genome-wide genealogy estimation for thousands of samples. *Nat. Genet.* 51(9):1321–29
85. Schiffels S, Durbin R. 2014. Inferring human population size and separation history from multiple genome sequences. *Nat. Genet.* 46(8):919–25
86. Terhorst J, Kamm JA, Song YS. 2017. Robust and scalable inference of population history from hundreds of unphased whole genomes. *Nat. Genet.* 49(2):303–9
87. Waples RS. 1989. A generalized approach for estimating effective population size from temporal changes in allele frequency. *Genetics* 121(2):379–91
88. Hollenbeck CM, Portnoy DS, Gold JR. 2016. A method for detecting recent changes in contemporary effective population size from linkage disequilibrium at linked and unlinked loci. *Heredity* 117(4):207–16
89. Barbato M, Orozco-terWengel P, Tapio M, Bruford MW. 2015. SNeP: a tool to estimate trends in recent effective population size trajectories using genome-wide SNP data. *Front. Genet.* 6:109
90. Santiago E, Novo I, Pardiñas AF, Saura M, Wang J, Caballero A. 2020. Recent demographic history inferred by high-resolution analysis of linkage disequilibrium. *Mol. Biol. Evol.* 37(12):3642–53
91. Waples RK, Larson WA, Waples RS. 2016. Estimating contemporary effective population size in non-model species using linkage disequilibrium across thousands of loci. *Heredity* 117(4):233–40
92. Browning SR, Browning BL. 2015. Accurate non-parametric estimation of recent effective population size from segments of identity by descent. *Am. J. Hum. Genet.* 97(3):404–18
93. Palamara PF, Lencz T, Darvasi A, Pe'er I. 2012. Length distributions of identity by descent reveal fine-scale demographic history. *Am. J. Hum. Genet.* 91(5):809–22
94. Kirin M, McQuillan R, Franklin CS, Campbell H, McKeigue PM, Wilson JF. 2010. Genomic runs of homozygosity record population history and consanguinity. *PLOS ONE* 5(11):e13996
95. Gutenkunst RN, Hernandez RD, Williamson SH, Bustamante CD. 2009. Inferring the joint demographic history of multiple populations from multidimensional SNP frequency data. *PLOS Genet.* 5(10):e1000695
96. Wakeley J. 2009. *Coalescent Theory: An Introduction*. Greenwood Village, CO: Roberts & Co. Publ.
97. Wallberg A, Han F, Wellhagen G, Dahle B, Kawata M, et al. 2014. A worldwide survey of genome sequence variation provides insight into the evolutionary history of the honeybee *Apis mellifera*. *Nat. Genet.* 46(10):1081–88
98. Cao L-J, Song W, Chen J-C, Fan X-L, Hoffmann AA, Wei S-J. 2022. Population genomic signatures of the oriental fruit moth related to the Pleistocene climates. *Commun. Biol.* 5:142
99. Ortego J, Céspedes V, Millán A, Green AJ. 2021. Genomic data support multiple introductions and explosive demographic expansions in a highly invasive aquatic insect. *Mol. Ecol.* 30(17):4189–203
100. McCoy RC, Garud NR, Kelley JL, Boggs CL, Petrov DA. 2014. Genomic inference accurately predicts the timing and severity of a recent bottleneck in a nonmodel insect population. *Mol. Ecol.* 23(1):136–50
101. You M, Ke F, You S, Wu Z, Liu Q, et al. 2020. Variation among 532 genomes unveils the origin and evolutionary history of a global insect herbivore. *Nat. Commun.* 11:2321
102. Elfekih S, Etter P, Tay WT, Fumagalli M, Gordon K, et al. 2018. Genome-wide analyses of the *Bemisia tabaci* species complex reveal contrasting patterns of admixture and complex demographic histories. *PLOS ONE* 13(1):e0190555
103. Soberon J. 1999. Linking biodiversity information sources. *Trends Ecol. Evol.* 14(7):291
104. Gilbert MTP, Moore W, Melchior L, Worobey M. 2007. DNA extraction from dry museum beetles without conferring external morphological damage. *PLOS ONE* 2(3):e272
105. Grewe F, Kronforst MR, Pierce NE, Moreau CS. 2021. Museum genomics reveals the Xerces blue butterfly (*Glaucopsyche xerces*) was a distinct species driven to extinction. *Biol. Lett.* 17(7):20210123
106. Díez-del-Molino D, Sánchez-Barreiro F, Barnes I, Gilbert MTP, Dalén L. 2018. Quantifying temporal genomic erosion in endangered species. *Trends Ecol. Evol.* 33(3):176–85
107. Gauthier J, Pajkovic M, Neuenschwander S, Kaila L, Schmid S, et al. 2020. Museomics identifies genetic erosion in two butterfly species across the 20th century in Finland. *Mol. Ecol. Resour.* 20(5):1191–205

82. First of several methods using the sequentially Markovian coalescent to infer historical variation in N_e using genome variation data.

106. Insightful review into the use of genome data from historical samples to assess conservation status.

108. Parejo M, Wragg D, Henriques D, Charrière J-D, Estonba A. 2020. Digging into the genomic past of Swiss honey bees by whole-genome sequencing museum specimens. *Genome Biol. Evol.* 12(12):2535–51
109. Kimura M, Maruyama T, Crow JF. 1963. The mutation load in small populations. *Genetics* 48:1303–12
110. Henn BM, Botigué LR, Bustamante CD, Clark AG, Gravel S. 2015. Estimating the mutation load in human genomes. *Nat. Rev. Genet.* 16(6):333–43
111. Klopfstein S, Currat M, Excoffier L. 2006. The fate of mutations surfing on the wave of a range expansion. *Mol. Biol. Evol.* 23(3):482–90
112. Cingolani P, Platts A, Wang LL, Coon M, Nguyen T, et al. 2012. A program for annotating and predicting the effects of single nucleotide polymorphisms, SnpEff: SNPs in the genome of *Drosophila melanogaster* strain w^{1118}; iso-2; iso-3. *Fly* 6(2):80–92
113. Cooper GM, Stone EA, Asimenos G, NISC Comp. Seq. Program, Green ED, et al. 2005. Distribution and intensity of constraint in mammalian genomic sequence. *Genome Res.* 15(7):901–13
114. Kumar P, Henikoff S, Ng PC. 2009. Predicting the effects of coding non-synonymous variants on protein function using the SIFT algorithm. *Nat. Protoc.* 4(7):1073–81
115. Adzhubei IA, Schmidt S, Peshkin L, Ramensky VE, Gerasimova A, et al. 2010. A method and server for predicting damaging missense mutations. *Nat. Methods* 7(4):248–49
116. Kleinman-Ruiz D, Lucena-Perez M, Villanueva B, Fernández J, Saveljev AP, et al. 2022. Purging of deleterious burden in the endangered Iberian lynx. *PNAS* 119(11):e2110614119
117. van der Valk T, de Manuel M, Marques-Bonet T, Guschanski K. 2021. Estimates of genetic load suggest frequent purging of deleterious alleles in small populations. bioRxiv. 696831. **https://doi.org/10.1101/696831**
118. Pracana R, Burns R, Hammond RL, Haller BC, Wurm Y. 2022. Individual-based modeling of genome evolution in haplodiploid organisms. *Genome Biol. Evol.* 14(5):evac062
119. Ross KG, Fletcher DJC. 1986. Diploid male production: a significant colony mortality factor in the fire ant *Solenopsis invicta* (Hymenoptera: Formicidae). *Behav. Ecol. Sociobiol.* 19(4):283–91
120. Mattila ALK, Duplouy A, Kirjokangas M, Lehtonen R, Rastas P, Hanski I. 2012. High genetic load in an old isolated butterfly population. *PNAS* 109(37):E2496–505
121. Lenancker P, Hoffmann BD, Tay WT, Lach L. 2019. Strategies of the invasive tropical fire ant (*Solenopsis geminata*) to minimize inbreeding costs. *Sci. Rep.* 9(1):4566
122. Zayed A, Constantin ŞA, Packer L. 2007. Successful biological invasion despite a severe genetic load. *PLOS ONE* 2(9):e868
123. Hebert PDN, Cywinska A, Ball SL, deWaard JR. 2003. Biological identifications through DNA barcodes. *Proc. R. Soc. Lond. B* 270(1512):313–21
124. Nichols R. 2001. Gene trees and species trees are not the same. *Trends Ecol. Evol.* 16(7):358–64
125. Bickford D, Lohman DJ, Sodhi NS, Ng PKL, Meier R, et al. 2007. Cryptic species as a window on diversity and conservation. *Trends Ecol. Evol.* 22(3):148–55
126. Hebert PDN, Penton EH, Burns JM, Janzen DH, Hallwachs W. 2004. Ten species in one: DNA barcoding reveals cryptic species in the neotropical skipper butterfly *Astraptes fulgerator*. *PNAS* 101(41):14812–17
127. Christmas MJ, Jones JC, Olsson A, Wallerman O, Bunikis I, et al. 2021. Genetic barriers to historical gene flow between cryptic species of Alpine bumblebees revealed by comparative population genomics. *Mol. Biol. Evol.* 38(8):3126–43
128. Christmas MJ, Jones JC, Olsson A, Wallerman O, Bunikis I, et al. 2022. A genomic and morphometric analysis of alpine bumblebees: ongoing reductions in tongue length but no clear genetic component. *Mol. Ecol.* 31(4):1111–27
129. Sousa V, Hey J. 2013. Understanding the origin of species with genome-scale data: modelling gene flow. *Nat. Rev. Genet.* 14(6):404–14
130. Heliconius Genome Consort. 2012. Butterfly genome reveals promiscuous exchange of mimicry adaptations among species. *Nature* 487(7405):94–98
131. Miles A, Harding NJ, Bottà G, Clarkson CS, Antão T, et al. 2017. Genetic diversity of the African malaria vector *Anopheles gambiae*. *Nature* 552(7683):96–100
132. Galtier N. 2019. Delineating species in the speciation continuum: a proposal. *Evol. Appl.* 12(4):657–63

110. Detailed review of the population genetic forces influencing genetic load and how it can be estimated.

133. Hey J, Waples RS, Arnold ML, Butlin RK, Harrison RG. 2003. Understanding and confronting species uncertainty in biology and conservation. *Trends Ecol. Evol.* 18(11):597–603
134. Redding DW, Mooers AØ. 2006. Incorporating evolutionary measures into conservation prioritization. *Conserv. Biol.* 20(6):1670–78
135. Sistri G, Menchetti M, Santini L, Pasquali L, Sapienti S, et al. 2022. The isolated *Erebia pandrose* Apennine population is genetically unique and endangered by climate change. *Insect Conserv. Divers.* 15(1):136–48
136. Podsiadlowski L, Tunström K, Espeland M, Wheat CW. 2021. The genome assembly and annotation of the Apollo butterfly *Parnassius apollo*, a flagship species for conservation biology. *Genome Biol Evol.* 13(8):evab122
137. Dhuyvetter H, Gaublomme E, Verdyck P, Desender K. 2005. Genetic differentiation among populations of the salt marsh beetle *Pogonus littoralis* (Coleoptera: Carabidae): a comparison between Atlantic and Mediterranean populations. *J. Hered.* 96(4):381–87
138. Crozier L, Dwyer G. 2006. Combining population-dynamic and ecophysiological models to predict climate-induced insect range shifts. *Am. Nat.* 167(6):853–66
139. Broquet T, Petit EJ. 2009. Molecular estimation of dispersal for ecology and population genetics. *Annu. Rev. Ecol. Evol. Syst.* 40:193–216
140. Waldvogel A-M, Feldmeyer B, Rolshausen G, Exposito-Alonso M, Rellstab C, et al. 2020. Evolutionary genomics can improve prediction of species' responses to climate change. *Evol. Lett.* 4(1):4–18
141. Savolainen O, Lascoux M, Merilä J. 2013. Ecological genomics of local adaptation. *Nat. Rev. Genet.* 14(11):807–20
142. Novembre J, Di Rienzo A. 2009. Spatial patterns of variation due to natural selection in humans. *Nat. Rev. Genet.* 10:745–55
143. Coop G, Witonsky D, Rienzo AD, Pritchard JK. 2010. Using environmental correlations to identify loci underlying local adaptation. *Genetics* 185(4):1411–23
144. Günther T, Coop G. 2013. Robust identification of local adaptation from allele frequencies. *Genetics* 195(1):205–20
145. Frichot E, Schoville SD, Bouchard G, François O. 2013. Testing for associations between loci and environmental gradients using latent factor mixed models. *Mol. Biol. Evol.* 30(7):1687–99
146. Capblancq T, Forester BR. 2021. Redundancy analysis: a Swiss army knife for landscape genomics. *Methods Ecol. Evol.* 12(12):2298–309
147. Adrion JR, Hahn MW, Cooper BS. 2015. Revisiting classic clines in *Drosophila melanogaster* in the age of genomics. *Trends Genet.* 31(8):434–44
148. Turner TL, Levine MT, Eckert ML, Begun DJ. 2008. Genomic analysis of adaptive differentiation in *Drosophila melanogaster*. *Genetics* 179(1):455–73
149. Machado HE, Bergland AO, Taylor R, Tilk S, Behrman E, et al. 2021. Broad geographic sampling reveals the shared basis and environmental correlates of seasonal adaptation in *Drosophila*. *eLife* 10:e67577
150. Balanyá J, Oller JM, Huey RB, Gilchrist GW, Serra L. 2006. Global genetic change tracks global climate warming in *Drosophila subobscura*. *Science* 313(5794):1773–75
151. Calfee E, Agra MN, Palacio MA, Ramírez SR, Coop G. 2020. Selection and hybridization shaped the rapid spread of African honey bee ancestry in the Americas. *PLOS Genet.* 16(10):e1009038
152. Wallberg A, Schöning C, Webster MT, Hasselmann M. 2017. Two extended haplotype blocks are associated with adaptation to high altitude habitats in East African honey bees. *PLOS Genet.* 13(5):e1006792
153. Christmas MJ, Wallberg A, Bunikis I, Olsson A, Wallerman O, Webster MT. 2019. Chromosomal inversions associated with environmental adaptation in honeybees. *Mol. Ecol.* 28(6):1358–74
154. Montero-Mendieta S, Tan K, Christmas MJ, Olsson A, Vilà C, et al. 2019. The genomic basis of adaptation to high-altitude habitats in the eastern honey bee (*Apis cerana*). *Mol. Ecol.* 28(4):746–60
155. Pimsler ML, Oyen KJ, Herndon JD, Jackson JM, Strange JP, et al. 2020. Biogeographic parallels in thermal tolerance and gene expression variation under temperature stress in a widespread bumble bee. *Sci. Rep.* 10(1):17063

140. Stimulating review of the ways population genomics can be used to predict species response to climate change incorporating adaptive potential.

156. Chen Y, Liu Z, Régnière J, Vasseur L, Lin J, et al. 2021. Large-scale genome-wide study reveals climate adaptive variability in a cosmopolitan pest. *Nat. Commun.* 12:7206
157. Gamboa M, Watanabe K. 2019. Genome-wide signatures of local adaptation among seven stoneflies species along a nationwide latitudinal gradient in Japan. *BMC Genom.* 20:84
158. Dudaniec RY, Yong CJ, Lancaster LT, Svensson EI, Hansson B. 2018. Signatures of local adaptation along environmental gradients in a range-expanding damselfly (*Ischnura elegans*). *Mol. Ecol.* 27(11):2576–93
159. Fouet C, Atkinson P, Kamdem C. 2018. Human interventions: driving forces of mosquito evolution. *Trends Parasitol.* 34(2):127–39
160. Bass C, Denholm I, Williamson MS, Nauen R. 2015. The global status of insect resistance to neonicotinoid insecticides. *Pestic. Biochem. Physiol.* 121:78–87
161. Straub L, Strobl V, Neumann P. 2020. The need for an evolutionary approach to ecotoxicology. *Nat. Ecol. Evol.* 4:895
162. Pélissié B, Chen YH, Cohen ZP, Crossley MS, Hawthorne DJ, et al. 2022. Genome resequencing reveals rapid, repeated evolution in the Colorado potato beetle. *Mol. Biol. Evol.* 39(2):msac016
163. Kiani M, Fu Z, Szczepaniec A. 2022. ddRAD sequencing identifies pesticide resistance-related loci and reveals new insights into genetic structure of *Bactericera cockerelli* as a plant pathogen vector. *Insects* 13(3):257
164. Singh KS, Cordeiro EMG, Troczka BJ, Pym A, Mackisack J, et al. 2021. Global patterns in genomic diversity underpinning the evolution of insecticide resistance in the aphid crop pest *Myzus persicae*. *Commun. Biol.* 4:847
165. Valencia-Montoya WA, Elfekih S, North HL, Meier JI, Warren IA, et al. 2020. Adaptive introgression across semipermeable species boundaries between local *Helicoverpa zea* and invasive *Helicoverpa armigera* moths. *Mol. Biol. Evol.* 37(9):2568–83
166. Hou Z, Wei C. 2019. De novo comparative transcriptome analysis of a rare cicada, with identification of candidate genes related to adaptation to a novel host plant and drier habitats. *BMC Genom.* 20:182
167. Simon J-C, d'Alençon E, Guy E, Jacquin-Joly E, Jaquiéry J, et al. 2015. Genomics of adaptation to host-plants in herbivorous insects. *Brief. Funct. Genom.* 14(6):413–23
168. Oppenheim SJ, Gould F, Hopper KR. 2012. The genetic architecture of a complex ecological trait: host plant use in the specialist moth, *Heliothis subflexa*. *Evolution* 66(11):3336–51
169. Caillaud MC, Via S. 2012. Quantitative genetics of feeding behavior in two ecological races of the pea aphid, *Acyrthosiphon pisum*. *Heredity* 108(3):211–18
170. Soria-Carrasco V, Gompert Z, Comeault AA, Farkas TE, Parchman TL, et al. 2014. Stick insect genomes reveal natural selection's role in parallel speciation. *Science* 344(6185):738–42
171. Carvell C, Bourke AFG, Dreier S, Freeman SN, Hulmes S, et al. 2017. Bumblebee family lineage survival is enhanced in high-quality landscapes. *Nature* 543(7646):547–49
172. Traynor KS, Mondet F, de Miranda JR, Techer M, Kowallik V, et al. 2020. *Varroa destructor*: a complex parasite, crippling honey bees worldwide. *Trends Parasitol.* 36(7):592–606
173. Mondet F, Beaurepaire A, McAfee A, Locke B, Alaux C, et al. 2020. Honey bee survival mechanisms against the parasite *Varroa destructor*: a systematic review of phenotypic and genomic research efforts. *Int. J. Parasitol.* 50(6):433–47
174. Broeckx BJG, De Smet L, Blacquière T, Maebe K, Khalenkow M, et al. 2019. Honey bee predisposition of resistance to ubiquitous mite infestations. *Sci. Rep.* 9:7794
175. Wood TJ, Michez D, Paxton RJ, Drossart M, Neumann P, et al. 2020. Managed honey bees as a radar for wild bee decline? *Apidologie* 51(6):1100–16
176. Sackton TB. 2019. Comparative genomics and transcriptomics of host-pathogen interactions in insects: evolutionary insights and future directions. *Curr. Opin. Insect Sci.* 31:106–13
177. Des Roches S, Pendleton LH, Shapiro B, Palkovacs EP. 2021. Conserving intraspecific variation for nature's contributions to people. *Nat. Ecol. Evol.* 5:574–82
178. Wellenreuther M, Bernatchez L. 2018. Eco-evolutionary genomics of chromosomal inversions. *Trends Ecol. Evol.* 33(6):427–40
179. Rellstab C, Dauphin B, Exposito-Alonso M. 2021. Prospects and limitations of genomic offset in conservation management. *Evol. Appl.* 14(5):1202–12

180. Fuller ZL, Mocellin VJL, Morris LA, Cantin N, Shepherd J, et al. 2020. Population genetics of the coral *Acropora millepora*: toward genomic prediction of bleaching. *Science* 369(6501):eaba4674

181. Exposito-Alonso M, Vasseur F, Ding W, Wang G, Burbano HA, Weigel D. 2018. Genomic basis and evolutionary potential for extreme drought adaptation in *Arabidopsis thaliana*. *Nat. Ecol. Evol.* 2(2):352–58

182. Bay RA, Harrigan RJ, Underwood VL, Gibbs HL, Smith TB, Ruegg K. 2018. Genomic signals of selection predict climate-driven population declines in a migratory bird. *Science* 359(6371):83–86

183. Beng KC, Tomlinson KW, Shen XH, Surget-Groba Y, Hughes AC, et al. 2016. The utility of DNA metabarcoding for studying the response of arthropod diversity and composition to land-use change in the tropics. *Sci. Rep.* 6(1):24965

184. Corlett RT. 2017. A bigger toolbox: biotechnology in biodiversity conservation. *Trends Biotechnol.* 35(1):55–65

180. Important example of the use of genomics to predict response to climate change.

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Contents

Errata

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