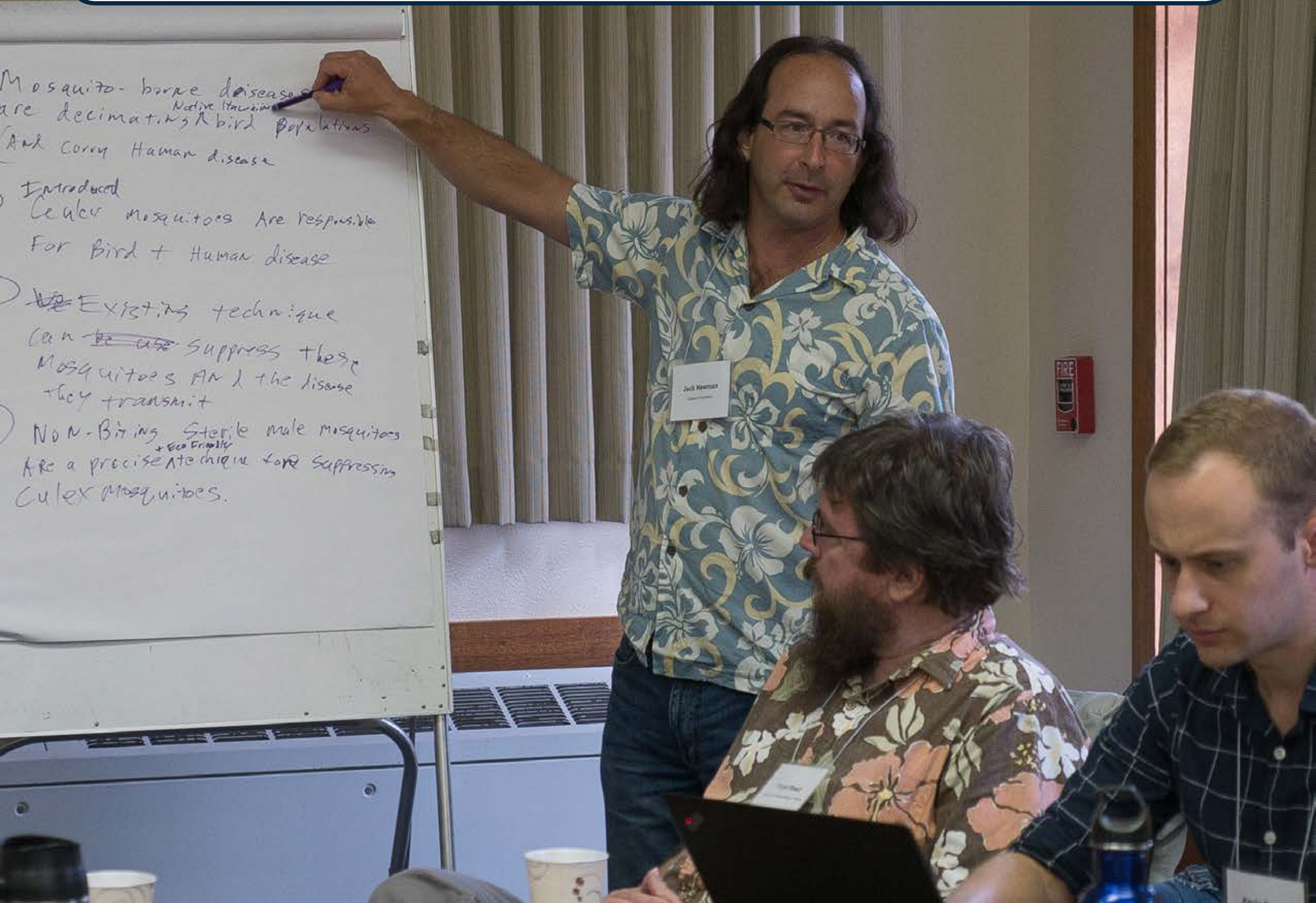
To Restore a Mosquito-Free Hawai‘i

Summary Report of the Workshop to Formulate Strategic Solutions for a “Mosquito-Free Hawai‘i”

A workshop was convened on September 6-7, 2016, to seek strategic solutions to eliminate mosquito-borne diseases affecting humans and wildlife. Workshop participants ranged from experts in mosquitoes and mosquito-borne pathogens to local leaders, public health and wildlife specialists. The discussions focused on novel technologies to **transform, suppress and ideally eliminate alien mosquito vectors from the Hawaiian Islands using an integrative systems thinking approach**. Attendees concluded that broad support to engage the public, develop the science and put resources to work on locally appropriate solutions is critical to combat serious threats of mosquito-transmitted diseases to protect both Hawai'i's public health and unique biodiversity. This white paper is a summary of the discussions of the workshop.



On the cover: *Aedes aegypti*, first introduced to the Hawaiian Islands after 1882, this invasive mosquito can transmit dengue, chikugunya and Zika virus.

To Restore a Mosquito-Free Hawai‘i

Summary:

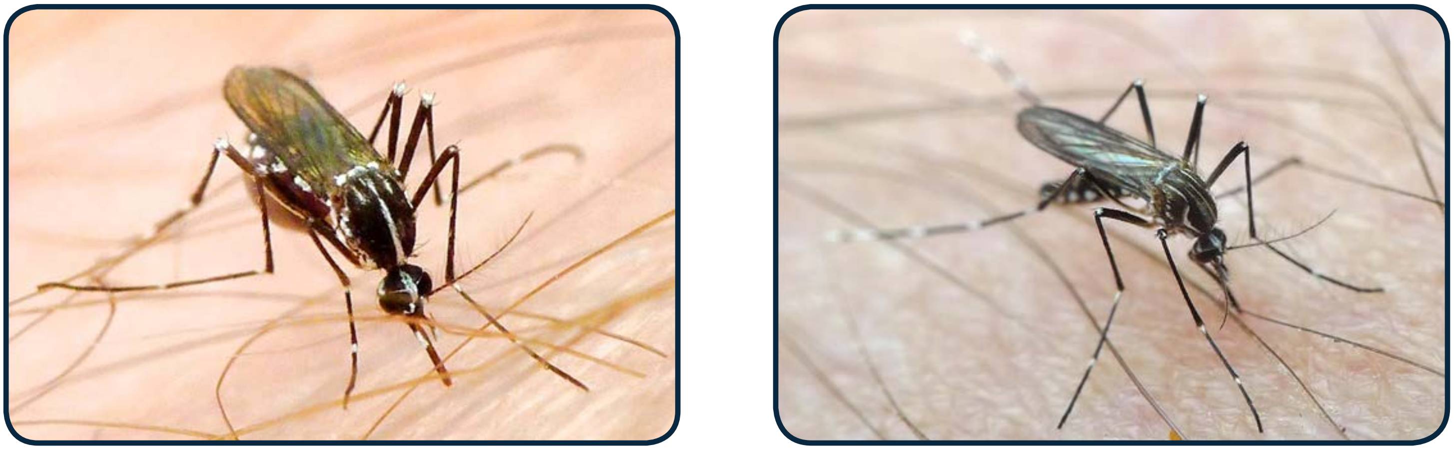
- Mosquitoes are non-native to the Hawaiian Islands.
- Mosquito-borne diseases are decimating native Hawaiian birds and threaten human health.
- There are new solutions to suppress or eliminate mosquitoes at an island-wide scale.
- A partnership with an engaged public, local experts, and a supportive government will be necessary to capitalize on this opportunity.
- For the first time, a path forward to re-establish a “mosquito-free” Hawai‘i is achievable.

Abstract:

Introduced mosquito species transmit diseases that threaten Hawai‘i’s public health, native forest birds, culture and economy. These existing mosquito-borne diseases, combined with impending threats of novel pathogens, have galvanized interest in new techniques to combat mosquitoes in Hawai‘i. Several targeted and effective strategies for mosquito suppression are currently available, and in five to ten years, more advanced tools may be available to completely restore a mosquito-free Hawai‘i.

Introduction:

Mosquitoes were introduced to Hawai‘i in the early 1800’s[1]. Six non-native mosquito species have become established since then, including two serious vectors of human diseases that threaten health, quality of life and the economy, as well as one vector of avian diseases that has contributed to the decline or extinction of many of Hawai‘i’s iconic native forest birds[2].



Mosquito species *Aedes albopictus* (L) *and Aedes aegypti* (R) can both transmit dengue, chikungunya, and Zika virus - Photos: (c) Durrell D. Kapan

The presence of mosquitoes in Hawai‘i represents a persistent and serious threat to public health, as well as to the economy and ecosystems. Diseases such as chikungunya, dengue, and yellow fever affect hundreds of millions of people worldwide, causing debilitating symptoms and sometimes death[3]. More recently, the Zika virus began to spread through the Americas, causing birth defects and neurological disorders[4]. These human diseases are transmitted by two mosquitoes, the yellow fever mosquito (*Aedes aegypti*) and the Asian tiger mosquito (*Aedes albopictus*), natives of Africa and Asia respectively. Both of these species have invaded Hawai‘i[1] and are responsible for sporadic outbreaks of imported dengue fever[5,6]. Similarly, either of these two species could sustain a Zika virus outbreak sparked by the arrival of an infected traveler[7]. Additionally, the Southern house mosquito (*Culex quinquefasciatus*) transmits avian malaria parasite and avian pox virus, major factors in the extinction of more than half of Hawai‘i's honeycreepers. The Southern house mosquito can also transmit West Nile virus which has not yet reached the islands[8]. This mosquito and the pathogens it carries threaten imminent extinction of most of the remaining 17 species of these unique birds that are found nowhere else on Earth[9].

Standard mosquito control methods cannot permanently suppress or eradicate mosquitoes in Hawai‘i. They are too costly, labor intensive, and often employ non-specific pesticides all of which are not effective or appropriate in rural and especially remote roadless forests where disease-sensitive native birds live. However, novel approaches offer new hope to control and even eliminate mosquitoes in Hawai‘i. Recent dengue outbreaks, combined with the threat of a local Zika virus epidemic, highlight Hawai‘i's vulnerability to mosquito-borne pathogens and have galvanized efforts to look beyond standard methods to minimize the risk of mosquito-borne diseases in the islands. Removing mosquitoes from the Hawaiian Islands would eliminate the threat of vector-borne diseases that currently impact human and native forest bird populations.

Mosquitoes in Hawai‘i Workshop: Novel approaches to confront mosquito vectors and mosquito-borne pathogens in the Hawaiian Islands

With the support of Hawai‘i County Mayor Billy Kenoi, a group of biologists, biotechnology experts, wildlife managers, and public health specialists gathered at Hawai‘i Volcanoes National Park on September 6 & 7, 2016, to discuss possible solutions to the problem of invasive mosquitoes in Hawai‘i. The following summarizes the discussion of mosquito-borne diseases in Hawai‘i and methods to control them by suppressing or eliminating mosquitoes at the landscape scale.

Mosquitoes are not native to the Hawaiian Islands and transmit non-native pathogens: Prior to the arrival of European ships and trade, the Hawaiian Islands had no native mosquitoes[1]! The first invasive species, the Southern house mosquito (*Culex quinquefasciatus*), was introduced around 1826 when sailors drained their water barrels on Maui[10]. Subsequently, the yellow fever mosquito (*Aedes aegypti*) and the Asian tiger mosquito (*Aedes albopictus*) were introduced between 1892 and 1900. Soon after their introduction, the Southern House Mosquito spread avian malaria and avian pox to Hawai‘i's unique forest birds, and the yellow fever and Asian tiger mosquitoes spread dengue fever to people. During the next century, three additional mosquito species were introduced to Hawai‘i, but they are not known to be vectors of pathogens detrimental to humans or Hawai‘i's native wildlife[1,11].

To Restore a Mosquito-Free Hawai'i

Mosquito-borne pathogens threaten the health of all people living in or visiting Hawai'i: The dengue virus hit Hawai'i less than a decade after the introduction of *Aedes aegypti* and *Aedes albopictus*. Over 30,000 people contracted dengue fever in 1903[12]. Since then, at least four additional outbreaks have occurred, including outbreaks on O'ahu (2001-02, 2011) and most recently, on Hawai'i Island (winter of 2015 and spring of 2016) with over 260 confirmed dengue cases [6,13]. With increases in travel, population size and mosquito density, people in Hawai'i can expect mosquito-borne illnesses such as dengue to rise in the future. Additionally, viruses new to Hawai'i such as chikungunya, West Nile, and Zika could rapidly spread through the immunologically-naive human population of Hawai'i because they are easily transmitted by mosquito species already present.

Mosquito-borne pathogens are decimating Hawai'i's vulnerable native forest birds: Due to the extreme isolation of the Hawaiian Islands, Hawai'i's native landbirds have the highest percentage of unique *endemic* species (98%) in the world[14]. These birds play important ecological roles and are also highly significant in Hawaiian culture[2]. While Hawaiian native forest birds are threatened by habitat loss, habitat degradation from invasive plants and invertebrates, introduced predators and competitors, it is widely accepted that introduced avian malaria and avian pox virus are responsible for ongoing range contractions and declining populations of many of these species. With no prior exposure or natural immunity, the native birds are highly susceptible to these non-native pathogens transmitted by the Southern house mosquito (*Culex quinquefasciatus*). Prior to the introduction of this mosquito and the pathogens it transmits, there were at least 50 native forest bird species in the main Hawaiian Islands. *More than 50% of these bird species have gone extinct, and more than half of those that still remain are currently on the brink of extinction, in large part because of mosquito-borne avian malaria and pox*[15]. As global temperatures rise, mosquitoes and the diseases they carry are moving into higher elevation forests, causing rapid population declines in many of the surviving bird species, including 'I'iwi (*Drepanis coccinea*), 'Akikiki (*Oreomystis bairdi*), 'Akeke'e (*Loxops caeruleirostris*), 'Anianiau (*Hemignathus parvus*) and Kaua'i 'Amakihi (*Chlorodrepanis stejnegeri*)[16]. The disease-cycle in bird populations can only be broken by suppressing or eradicating mosquitoes. Unless this action is taken , avian malaria and avian pox are expected to spread to all remaining disease-free forest habitats and lead to the extinction of the rarest of Hawai'i's unique honeycreepers[16–18].

'Apapane (*Himatione sanguinea*), a crimson red Hawaiian honeycreeper, being bitten by the alien invasive mosquito *Culex quinquefasciatus*. Photo: (c) Jack Jeffrey

In summary, non-native mosquitoes in Hawai'i have caused human disease epidemics and the severe loss of biodiversity. If mosquitoes remain unchecked, they will continue to negatively impact human health and cause the extinction of most of the remaining Hawaiian forest bird species.

Potential Solutions:

Conventional methods will not solve the mosquito problem: The approaches most often employed for mosquito control in urban areas cannot address the unique challenges of Hawai'i at the landscape scale. The cornerstone of mosquito control, source reduction, aims to limit the watery habitats where mosquitoes breed by eliminating refuse, used tires, covering cisterns, cleaning gutters, and emptying other containers[19,20]. Insecticides are often used during health emergencies to try to knock down potentially infected adults that are transmitting a disease, but factors such as vegetation make this problematic in Hawai'i[19,20]. Other mosquito control tools include biological insecticides developed from the bacterium *Bacillus thuringiensis* (Bti), which are applied to watery breeding habitats to eliminate mosquito larvae[21]. These approaches can be somewhat effective when used to control the yellow-fever mosquito (*Aedes aegypti*), when found breeding in accessible urban habitats[22]. However, these methods are not feasible for landscape level control of mosquito species that can breed in rural, forested and wilderness habitats in Hawai‘i. Broad application of insecticides to forested areas inhabited by native birds is not feasible not only because it would be logistically difficult and expensive, but also because it would have undesirable effects on native species, watersheds and human health [23]. Another control option is to place traps with chemicals that attract and kill females that seek water in which to lay eggs[5], known as the lethal ovitrap method[24]. This approach has been used during recent dengue outbreaks in Hawai‘i, and it can help control *Aedes aegypti* around homes and people[25.]. However, lethal ovitraps are impractical for broad landscape level application in forests and rural areas because a very large number of traps would need to be placed, monitored and maintained. Moreover, once chemicals degrade, the traps themselves can become mosquito breeding grounds.

We can use alternative methods to address the mosquito problem: A different class of methods solves many of the problems described above by targeting the mosquitoes directly using their own unique biology. New applications of the Sterile Insect Technique (SIT) provide the opportunity for the precise suppression of mosquitoes with no direct effects on other species and no negative impacts on human health[26]. In its simplest form, male mosquitoes are sterilized and released into the wild so that when they mate with females, they either produce no offspring or their offspring cannot effectively survive and reproduce. Over time, and with enough sterile male releases, fewer and fewer mosquitoes survive and breed, and eventually the mosquito population crashes. Importantly, male mosquitoes do not bite, and their release poses no health concerns. Notably, since sterile males die without successfully reproducing, these SIT methods are ‘self-limiting’ meaning the mosquitoes do not persist in the wild.

SIT was developed in the 1950s to eliminate agricultural pests in the United States[27]. This technique successfully eliminated screwworms, a livestock pest, from all of North and Central America, the island of Curaçao, and regions of Africa. SIT also has been used to eradicate the Mediterranean fruit fly in Mexico and California, the Oriental fruit fly and the melon fly in Okinawa, and to help control the tsetse fly in Africa[27].

Available SIT technologies: There are three types of self-limiting SIT that have been tested in the field and are now available to use individually or in combination to control or eliminate non-native mosquitoes with no direct non-target effects[28].

(i) Releases of male mosquitoes sterilized by irradiation: For the last 50 years, SIT has been achieved by sterilizing male insects with irradiation. Irradiated males are then released to seek out and mate with females of their own species. Because the males are sterile, any females they mate with will not produce offspring. With sufficient releases of sterile male mosquitoes, the wild population will eventually be reduced to a very low level or be locally eliminated[27]. Because irradiated males don't produce viable offspring and die after one to two weeks, this approach requires sustained releases of sterile males to maintain effective suppression. Hawai'i has an existing agricultural irradiation facility that can sterilize mosquitoes, making it possible to apply SIT to mosquito species in Hawai'i[29]. Although irradiation-based SIT has been successfully used for multiple agricultural pests such as the screwworm and medfly, irradiated mosquitoes do tend to have reduced fitness compared to wild-type males[30]. Specifically, the irradiation dose required to fully sterilize male mosquitoes can also cause the males to be less competitive for mates. Several laboratories are actively working to overcome this complication.

(ii) Releases of male mosquitoes carrying the bacterium *Wolbachia*: Suppression and elimination of mosquito populations can also be achieved by releasing male mosquitoes that carry insect specific bacteria called *Wolbachia*. Because these bacteria are highly specialized and cannot survive outside mosquito cells, they are completely harmless to humans and birds. Many different strains of *Wolbachia* are naturally found in about half of all insects[31], including those native to Hawai‘i[32]. In nature, *Wolbachia* are passed on from females to their offspring, but scientists can also introduce new strains of *Wolbachia* into insects in the laboratory. Various strains of *Wolbachia* have been successfully introduced into the yellow fever mosquito, the Asian tiger mosquito and the southern house mosquito in the laboratory, and it was discovered that these *Wolbachia* suppress the development of viruses like dengue, chikungunya, West-Nile and Zika in mosquito tissues[33,34]. *Wolbachia* can also work as a SIT known as the Incompatible Insect Technique (IIT)[35,36] through a mechanism called cytoplasmic incompatibility[30]. Namely, matings between male and female mosquitoes with different, incompatible strains of *Wolbachia* will fail to produce living embryos[30], so when many incompatible males are released to mate with local females, this causes mosquito populations to crash[36]. *Wolbachia* male-based IIT programs have shown progress in controlling local populations of *Aedes* and *Culex* mosquitoes around the globe[30,37,38] and this approach has received federal, state, and local approvals allowing field trials in California, Florida, and Kentucky[39]. These *Wolbachia*-male technologies could be readily adapted for populations of *Aedes aegypti*, *Aedes albopictus*, and *Culex quinquefasciatus* in Hawaii. Given that *Wolbachia* are passed only from mother to offspring, released males cannot spread the novel *Wolbachia*. This makes the *Wolbachia*-male method self-limiting, meaning novel *Wolbachia* cannot spread into the wild mosquito population. However, because laboratory females that carry novel *Wolbachia* can be accidentally released alongside males, sex separation is required to ensure only males are released[30]. Current sex separation techniques are not 100 percent effective, therefore they are the focus of intense research and development, along with continued work to au-

The Southern house mosquito, *Culex quinquefasciatus*, is a vector of avian malaria and avian pox tomate and reduce the costs associated with the mass rearing of mosquitoes[40].

(iii) Releases of irradiated male mosquitoes that carry *Wolbachia:* To overcome the issue of imperfect sex separation and accidental releases of females that carry novel *Wolbachia*, another approach has been developed. This approach combines the best aspects of methods from (i) and (ii) to reduce or eliminate mosquito populations. A much lower dose of radiation is required to sterilize female mosquitoes than males[41]. Thus, irradiating *Wolbachia*-infected mosquitoes can reliably sterilize the very small number of residual females that may be mixed with males intended for the release. At the same time, the *Wolbachia*-infected males mate with the wild-type females and affect their reproductive capacity as described in (ii) above[41]. This combined technique prevents accidental local establishment of the novel *Wolbachia* in mosquito populations. The combination approach has been used in a release of five million male mosquitoes per week in southern China, reducing local populations of *Aedes albopictus* by >90% (Zhiyong Xi pers. comm.). A similar method could be readily developed for local populations of *Wolbachia* and each invasive mosquito to achieve landscape level control.

(iv) Release of Self-Limiting male mosquitoes: A fourth method that is field-ready is the application of 'Self-Limiting' insects. The approach uses genetic technology to provide a means of preventing survival of the offspring of released males in the field, without the fitness reduction associated with methods that rely solely on irradiation. Males carrying edited genes are released into the field, where they seek and mate with females of their species, but they either do not produce offspring or their offspring die at immature stages (larvae and pupae)[26]. Because Self-Limiting males don't produce viable offspring, the edited gene does not persist in the environment. Like the other techniques, the Self-Limiting method also requires sustained releases to maintain effective control. The self-limiting strategy has demonstrated field success against *Aedes aegypti* by reducing target populations by >90% in several localities around the globe[42], has received a regulatory finding of no significant impact (FONSI) by the FDA[43], could be readily implemented to control this mosquito in Hawai'i, and can be applied to other important disease-transmitting mosquitoes such as *Aedes albopictus*[44].

New technology on the horizon: New genetic approaches for mosquito population suppression are being investigated under laboratory conditions. These differ fundamentally from SIT methods outlined above by employing a mechanism, termed gene drive, to increase the inheritance of particular genes in breeding populations of organisms[45,46]. By ensuring that they are always inherited, such gene drive systems can increase the frequency of specific traits, even if these don't benefit the organism. For example, one application might ensure that all mosquito offspring are male, or might cause infertility in females whenever both parents carry the drive system. Either way, natural mating will cause the change to spread through the local population, steadily decreasing the number of newly-hatched mosquitoes. In principle, this could allow permanent removal. Some potential long-term advantages of such approaches include many fewer releases, much lower cost, no direct impact on non-target species, and the ability to swiftly and cheaply eliminate any population that re-invades the islands[47,48]. Several milestones are absolutely necessary before society in general, and scientists specifically, could safely test gene drives to control mosquitoes in the wild. These include procedures to mitigate unanticipated outcomes during development as well as reliable methods of limiting the impact to a particular area or region.[48] Any project seeking to develop these systems must be fully transparent and engage in close consultation with communities in Hawai'i to be considered for future use[47].

To Restore a Mosquito-Free Hawai‘i

Issues:

Data needs: In order to consider and effectively deploy any of these methods, additional key information is needed to better inform stakeholders. Ecological data on *Culex quinquefasciatus* in Hawai‘i Volcanoes National Park and other rural and forested habitats in Hawai‘i[49–54], plus historic data and relatively recent vector control surveys for *Aedes aegypti* and *Aedes albopictus* provide an excellent beginning[5,55]. However, there is still a need for further information such as the baseline distribution, range of habitats, population structure and population sizes of each species of mosquito. This information is critical to assess the feasibility of various approaches and how they may scale to the landscape level. If a particular project is approved, ongoing monitoring will be needed to accurately assess progress towards suppressing or eliminating mosquitoes from Hawai‘i, and to detect any reinvasion of mosquitoes to areas once they have been removed.

Mosquito ecology and native species: Mosquitoes are not native to Hawai‘i, and any ecological role they may fill as prey, pollinators, or resource processors, will have originated recently. Therefore, native species are not likely to have become dependent upon them as a critical resource. Although adult mosquitoes could be potential food for Hawai‘i's native insectivores (‘ōpe‘ape‘a, the Hawaiian hoary bat; *Lasiurus semotus*[56,57], or the three endemic species of ‘elepaio, monarch flycatchers in the genus *Chasiempis*), they are not thought to form a significant fraction of these insectivores' diets due to their small body size compared to larger, more preferable prey items. Even if the removal of a particular mosquito species does not have a direct negative effect on a native species, it is important to understand potential indirect effects. Although mosquitoes are not native to Hawai‘i, further studies should be conducted to better understand the role mosquitoes play in Hawaiian ecosystems.

Community Engagement: Participants at the mosquito workshop in Hawai‘i Volcanoes National Park unanimously agreed that transparency, education, and community outreach are integral components of any landscape scale mosquito control aimed at protecting people's health and preventing forest bird extinctions. At the workshop, which was attended by several local leaders, numerous participants called for active community guidance of any proposals from the earliest stages. Achieving a mosquito-free Hawai‘i would require authentic and sustained engagement among local communities and a wide range of other stakeholders. Success will be unlikely without their unique knowledge and contributions. Therefore, it is essential that appropriate community engagement strategies are designed and implemented from the outset and sustained throughout[58].

Next steps: First and foremost is the question of how to involve all residents in determining the ecological and public health future for Hawai‘i. A forum is needed to hear from groups and communities that are most affected by mosquitoes. A broad coalition must be established to study the dimensions of the problem to collectively work towards sustainable solutions. A plan should be mapped out that can address both social and technical concerns related to these technologies. All planning must include relevant community input, and funding must be secured to accomplish this essential component of any mosquito control plan. Simultaneously, it will be necessary to devote additional resources to conduct further research and development of safe, targeted, efficient mosquito control technologies appropriate for Hawai‘i.

To Restore a Mosquito-Free Hawai‘i

Putting resources to work, engaging the public, and developing the science are vital first steps in order to halt the extinction of Hawai‘i's unique forest birds and to take measures to address the serious threats that mosquito-transmitted diseases pose to public health in Hawai‘i.

Conclusion:

Mosquito species introduced within the last two hundred years threaten Hawai‘i's public health, endemic forest birds, culture, and economy. The urgency of problems such as Zika and the imminent extinctions of several of Hawai‘i's forest birds have galvanized a critical mass of support to investigate the application of sterile insect techniques to re-establish a mosquito-free Hawai‘i. Mosquitoes that carry human diseases are a natural starting point to target for elimination or control with existing tools. Regional elimination of mosquitoes carrying bird diseases is also a feasible goal and is the best chance to avert the impending extinction of the endemic honeycreepers, ‘Akikiki, ‘Akeke‘e, ‘Anianiau, Kaua‘i ‘Amakihi. Several targeted and effective strategies for mosquito suppression are currently available, and in five to ten years, more advanced genetic tools may be available. Support of the residents of Hawai‘i will be critical to re-establish a mosquito-free Hawai‘i.

Unless immediate action is taken, people will continue to suffer from mosquito-borne diseases, and avian diseases will continue to threaten the existence of Hawai‘i's unique passerines.

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