

I am **OPPOSED** to this "Mosquito Suppression Environmental Assessment - December 2022" for the island of Maui. I demand that the State of Hawaii and its multi-agency partnership *Birds, Not Mosquitoes* complete a detailed, full scope **Environmental Impact Statement (EIS)** documenting the impacts to our native birds, wildlife, environment, and public health.

The State of Hawaii's planned "Mosquito Control Research Using *Wolbachia*-based Incompatible Insect Technique" project is promoted as a population control effort to save Hawaii's endangered native birds. The state has notified the public that the mosquito control strategy being implemented has decades of research behind it and is therefore safe. The reality is that the Sterile Insect Technique (SIT) that these decades of research are based on uses irradiation-induced sterility, and this is not the technique planned for use in this project. The Incompatible Insect Technique (IIT), based on *Wolbachia*-induced cytoplasmic incompatibility (a kind of male sterility), planned for use here in the islands is something entirely different. It is also a relatively new approach, and certainly has not been studied in Hawaii's unique ecosystems. Further, the biopesticide planned for use, "DQB Males," has not gone through the EPA registration process of scientific, legal, and administrative procedures through which the biopesticide would be examined for ingredients; site use; amount, frequency, and timing of use; and storage and disposal practices. Risk assessments to evaluate harms to humans, wildlife, fish, plants, endangered species, and non-target organisms have not been conducted. Potential contamination of surface water or ground water, leaching, runoff, and spray drift have not been evaluated.

This planned project comes with many risks to the land, birds, wildlife, and people of the Hawaiian Islands. While state agencies and wildlife officials are hoping this new strategy will prevent extinction of the native birds, it may very well cause their extinction. Some of the possible dangers include horizontal transmission of the introduced bacteria strain¹, increased pathogen infection in mosquitoes², irreversible evolutionary events¹, population replacement³ (lab-bred mosquitoes replacing existing wild mosquitoes), accidental release of lab-reared females³, creation of lab-strain females in the wild¹, horizontal gene transfer⁴, biopesticide drift, and mosquitoes becoming a better vector of avian malaria² and/or West Nile Virus⁵ (human and bird). Peer-reviewed studies document precedents for these concerns.

The Maui Environmental Assessment (EA) lists numerous potential impacts that require mitigation measures: wildland fire ignition by helicopters; spread of invasive weeds; noise-producing activities adversely affecting native wildlife; disturbances to native and special status plants and acceleration of erosion; noise disturbances and other impacts to special status wildlife species (including disturbances to nesting and roosting); adverse impacts within critical special status species habitats; transport and establishment of introduced invasive weeds and diseases/pathogens; disturbances of traditional cultural practices; threats to human health and safety; noise impacts on landowners, communities, wilderness, and sensitive environmental resources; noise and viewscape impacts on the visitor experience; impacts to wilderness character; threats to endangered nene and Hawaiian waterbirds; risks to Hawaiian forest birds and Hawaiian hoary bats from drone hovering and helicopter rotor wash; risks of breeding birds being flushed from active nests; disturbances of day roosting Hawaiian hoary bats; and risks of disturbing bat pup rearing. In the "Drone Release" section of the EA, it's noted that, "mosquitoes would likely be released in small biodegradable packages designed to open upon contact with the canopy or forest floor." The EA also states that, "these mosquito packages (dropped via aerial means) would result in an impact to the undeveloped quality of wilderness for as long as they remain in the environment (until they biodegrade)." The effects of this packaging on the environment and wildlife are not addressed.

The EA notes that the sound produced by each drone "is similar to loud highway noise," that "drone noise could possibly be loud enough to disrupt conversations," and that aircraft wildlife collisions could happen. The document states that "it is possible that a drone could inadvertently fly into a flock of birds." Helicopters planned for use in this project would be even louder, larger, more powerful, and more dangerous to birds and wildlife.

Environmental Justice is also a clear concern with this project. Per EPA documentation⁶, the "EPA seeks to achieve environmental justice, the fair treatment and meaningful involvement of any group, including minority



and/or low- income populations, in the development, implementation, and enforcement of environmental laws, regulations, and policies. To help address potential environmental justice issues, the Agency seeks information on any groups or segments of the population who, as a result of their location, cultural practices, or other factors, may have atypical or disproportionately high and adverse human health impacts or environmental effects from exposure to the pesticide(s) discussed in this document, compared to the general population."

In the EA's "Cultural Impact Assessment" section, seven Native Hawaiian lineal descendants and recognized cultural experts were interviewed. All expressed concerns about the impacts of the project, focused on the effects it could have on cultural resources and traditions, native birds, public health, wildlife, and our fragile ecosystems. Additional concerns include the experimental aspect of the project; the state's history of creating new problems by bringing in invasive species such as the mongoose; the sensitivity of the project area, with people depending on native flora and fauna for their livelihoods; impacts on other animals like 'opae (shrimp) and 'o'opu (goby fish) that live in streams; whether or not adequate studies or research have been done; residual effects on other insects; impacts on other islands from water sources connected through tides and currents; and the need to keep the public informed. The state's assessment concludes, "If the project and concerns about the use of this biocontrol discourage practitioners from conducting their traditional or customary practices, it would be an adverse effect to these cultural activities." As a result of their location, cultural practices, and other factors, **Native Hawaiians may have atypical or disproportionately high and adverse human health impacts and environmental effects from exposure to the biopesticide**.

A primary concern of this project is the potential for increased pathogen infection due to non-sexual horizontal transmission of the introduced *Wolbachia* strains between the introduced biopesticide mosquitoes and the existing "wild" mosquitoes. Adding to that concern are the diseases that the mosquito species planned for import into Hawaii transmit. The Southern House Mosquito (*Culex quinquefasciatus*) can transmit **avian malaria** parasitic disease to birds and **West Nile virus** to both birds and humans.

Horizontal transmission is defined as the spread of an infectious agent from one group or individual to another, directly or indirectly. In the case of the host arthropods (insects) and the infectious agent of *Wolbachia* bacteria, the horizontal transmission referenced here would be non-sexual. Imported *Wolbachia* bacterium strains involved in this project include wAlbA, wAlbB, and wPip4. These newly introduced strains (referred to here as "X") are not currently present within the corresponding *Culex quinquefasciatus* species of Hawaii's established mosquito population.

Tropical disease and vector expert Dr. Lorrin Pang has spoken as a private citizen about horizontal transmission, or "horizontal spread," of disease and the risks involved with this project. Pang has authored over 75 publications in peer-reviewed medical journals covering a broad range of studies such as malaria, dengue, rabies, rat lungworm, and COVID. He's been an advisor and voting member of the U.S. Congress Medical Research Program for the past several years, serving on committees for infectious diseases – many of which are mosquitoborne. From 1985-2005, he worked with the WHO and Walter Reed Institute's Malaria Program, focusing on global malaria control efforts through interventions combining diagnostics, chemotherapeutics, vector control, and vaccine development. As a public health leader on the islands, he has mitigated mosquito-borne illnesses – including dengue and Zika – for over two decades. Pang was honored for his life-saving intervention in Hawaii's dengue fever outbreak.

Dr. Pang has been compiling studies documenting horizontal *Wolbachia* bacterial spread, and he's concerned about the potential for significant adverse outcomes of the state's proposal:

"The intent to save rare birds is sound. If the project goes as planned, this would be a valuable tool for future interventions. However, with new life forms coming to the islands, there is too much potential for unexpected, dangerous, **irreversible 'evolutionary' events**. This is especially true when the new



organisms cannot be contained to their target ecosystem. Already there are published papers pointing out the real threat of **horizontal spread** of the novel *Wolbachia* beyond the male *Culex* mosquito. The papers cover two general areas – the widespread detection of *Wolbachia* across so many diverse types of insects, and more recently, the growing number of reports of mechanisms of how this might occur. First, we all must agree that unintended horizontal spread of Wpip4 (imported strain) to, say, female *Culex*, *Aedes* mosquitoes (human disease vectors), or other insect vectors of diseases would be a catastrophe, and probably irreversible. Hawaii has a bad history of invasive species entering and spreading unabated, including their spread of infectious diseases.

Proponents may be right that this intervention will save the native birds in the short-term, but long-term consequences to other island ecologies and to these same native birds may ultimately be detrimental. When one realizes the latter, the damage may be impossible to recall or repair, like the effect we've seen with so many other invasive species in Hawaii."

The safety assurances of the state's biopesticide project are based heavily on the premise that only male mosquitoes will be released. Because the males are infected with an incompatible bacteria strain, when they mate with existing wild females, the offspring are not viable. However, Dr. Pang points to a more recent study out of Singapore¹ describing *Wolbachia* bacteria strain "evolutionary associations" between mosquito hosts. The results of this mechanism widespread into diverse insect populations are not known. It may start with a few horizontal transfers to female mosquitoes. After that, the mating *Wolbachia*-X-compatible pair will quickly produce viable X offspring and spread the X bacteria strain (the term for this is "sweep"). If that were to happen here, the full capacity of those offspring to transmit disease would be unknown. This type of spread and sweep could also affect other insects, not just the targeted mosquito.

The combination of horizontal and vertical transmission dramatically contradicts the state's safety narrative. While the potential for accidental misidentification and release of lab-reared X-infected females (who bite and breed) has already been downplayed, the possibility of unintentionally producing these females in the wild has not been addressed at all. As Pang puts it,

"It is enough to say that the new *Wolbachia* strain can spread horizontally as a life form to other mosquitos (say *Aedes*, the vectors of human disease) and perhaps create that *Wolbachia* female *Culex* which everyone is bending over backwards to avoid via lab contamination."

Dr. Pang further points out that there is a big difference between the standard Sterile Insect Technique (SIT) strategies used in the past that were based on radiation or chemicals, and the relatively new Incompatible Insect Technique (IIT). The mathematical models may be similar for estimating threshold criteria to affect mosquito population dynamics, but standard methods of sterility are not bacterial life forms that might escape horizontally and amplify in other ecological niches. According to Pang,

"While sterility models can predict the thresholds needed to exterminate a species (in this case insects), the radiation sterility factor (standard SIT) does not behave the same as a life form (i.e., Wpip4 *Wolbachia* bacteria). There is very different modeling for the target insect – but more importantly, for the unintended groups to which the bacteria horizontally spread. How is this supposed to be self-contained? Horizontal spread has the potential to be a disaster that cannot be recalled. The bacterium is a life form, and you might not be able to turn back the clock by simply shutting off the male mosquito 'fountains.' "

The evidence of horizontal spread of *Wolbachia* shows that the bacteria go not only to sexual cells, but also to somatic cells (non-sexual cells of the body). *Wolbachia* can also live outside of intra-cellular systems for several months.⁷ Dr. Pang emphasizes two additional studies documenting widespread horizontal transmission of *Wolbachia*. The first focuses on predatory wasps spreading the bacteria through contaminated mouth parts when feeding serially on target insects such as aphids⁸. Pang calls for more research into which predators, like the



damselfly and dragonfly, sequentially feed on both male and female mosquitoes. This scenario might play out in either the predator of adults feeding on adult mosquitoes (X-infected and wild), or the X-infected predator of larva feeding on wild mosquito larva in common breeding sites. The second study looks at ant colonies spreading *Wolbachia* through the gastrointestinal (GI) tract when the ants feed on their fungus gardens.⁷ Pang asks an important question, "What about shared sugar feeding sites for X-infected male and wild adult male and female mosquitoes?" The sparser the sugar sites, the more communal interaction they will have. Dr. Pang finds these studies of horizontal transfer across species of insects worrisome, and says, "Even if this project achieved miraculous blocking of avian malaria to the native birds, what else would it do?"

To complicate matters more, the *Wolbachia* bacteria itself is parasitic, manipulating the reproductive biology of the host to increase its own transmission. Parasitic organisms can also alter the behavior of the hosts they live inside, and we just don't know how this might play out in our native bird habitats. Will the X-infected mosquitoes or their offspring be capable of moving up to even higher elevations? Would they be more aggressive? How would increased pathogen infection and elevated capacity for disease transmission factor in these scenarios?

Consider the example posed by the article "Parasites brainwash grasshoppers into death dive,"⁹ where a parasitic worm brainwashes the grasshopper host it invades to jump into water and commit suicide. The parasite accomplishes this by chemically influencing the grasshopper's brain, producing proteins which directly and indirectly affect the host's central nervous system. This causes an altering of the grasshopper's behavior so that it acts in a way it never usually would. Other parasites are noted as manipulators of their hosts' behavior, including "enslaver" fungi that make their insect hosts die perched in a position that favors the dispersal of spores by the wind. It is widely believed that *Wolbachia* bacteria is such a parasite (intracellular) that modifies the mosquito host's behaviors in ways we are only now beginning to understand.

In addition to the possibility of creating lab-strain females in the wild through horizontal transmission, there is also the issue of accidental release of misidentified females. Per the Department of Land and Natural Resources "Permit Application for Restricted Commodities into Hawaii" for the Southern House Mosquito *Culex quinquefasciatus*¹⁰:

"MosquitoMate and Verily will regularly sample release containers by releasing the contents into lab cages and then examining mosquito sex and number. There is an EPA reviewed value of 1 female release per 250,000 males with the MosquitoMate product. A similar value is likely to be estimated for *Culex quinquefasciatus* given that similar automation, engineering, and machine learning technology is being applied to sex sorting."

The EPA's "Emerging Mosquito Control Technologies" webpage¹¹ confirms these figures in the section titled, "Will this technology adversely affect human health or the environment?":

"Like all pesticides, MosquitoMate's technology is regulated under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA)...The expected accidental release rate of one *Wolbachia* infected female for every 250,000 infected males is considered negligible exposure to humans resulting in a negligible human health risk."

With posting of the EA, we've come to learn that the Maui project area covers **64,666 acres**, and that the state intends to release between 50 and 6,000 lab-bred mosquitoes per acre per treatment. Treatments would occur up to twice per week, amounting to up to **775,992,000 mosquitoes per week**. That is over 40 BILLION invasive biopesticide mosquitoes released per year just on the island of Maui!

With a release rate of one female for every 250,000 males, there could be up to 3,103 lab-strain-infected females released on the island per week. Just one accidental female mosquito released can produce 160,000 females in



her eight-week lifespan. This is a conservative estimate that takes into account a 50% mortality rate for offspring, a low-end of 100 eggs laid (the range is 100-300), an 80% emergence rate, an even sex ratio, and a first egg lay only (studies have found *Culex* females with two sperm packets, indicating two matings). One female *Culex quinquefasciatus* female can produce 20 living daughters in two weeks. Those daughters can produce 400 living granddaughters by four weeks. Those 400 living granddaughters can produce 8,000 living great-granddaughters by six weeks. Those great-granddaughters can produce 160,000 living great-granddaughters by the eighth week of the first female released.

These are not incompatible females. They will produce viable young when mated with the lab-reared males, and with any wild males that are not infected with the *Wolbachia* bacteria (not all *Culex* in the wild are infected with *Wolbachia*). If 3,103 lab-reared females are accidentally released per week, and each female can produce 160,000 more females, that amounts to potentially 496,480,000 (close to half a billion) accidental introduced-strain female mosquitoes within each eight-week life span of the initial accidental release scourge. These females bite and can spread disease. This is not addressed at all in the EA.

Population replacement can occur through mating of accidental lab-reared females and mating of wild mosquitoes who have become infected with the introduced *Wolbachia* bacteria strain through horizontal transmission. If the wild mosquitoes are replaced by the lab-strain-infected mosquitoes, the outcomes for our endangered native birds, public health, and our fragile ecosystems are unknown. What if the entire mosquito population becomes more capable of transmitting disease to birds, humans, and other wildlife?

These mosquito releases are planned indefinitely until the state finds another solution. This project intends to expand to Kauai next, and then throughout the islands. The EA notes an interest in establishing at least one **biopesticide lab here in Hawaii.** Federal level information¹² describes long-term plans for the islands, including lab research and development, "gene drives," "next generation tools," "synthetic biology control tools," "novel technology deployment," and "precision-guided Sterile Insect Technique (pgSIT)" (CRISPR technology). **"Genetic Modification of Forest Birds" using CRISPR-Cas9 gene editing** is also discussed in the EA. While "technology for this approach is not available for near-term implementation," development and deployment of this "tool" does appear to be a goal at the federal level. One of the labs that the state would be importing the biopesticide mosquitoes from, **Verily Life Sciences, is a subsidiary of Google**.

Per the "Understanding the Risks" section of the U.S. Department of the Interior Strategy document¹², "although used world-wide for human health, *Wolbachia* IIT is a novel tool for conservation purposes and **its degree of efficacy in remote forest landscapes is unknown**." This project is an experiment on Hawaii's people, wildlife, and 'aina. Not only is the extent of the risks involved undetermined, but the outcome itself of the planned goal is admittedly unknown. Because this is an experiment involving human disease vectors, the informed consent of the public is required. We do not consent.

Public testimony has shown that the people of Hawaii are overwhelmingly (over 75%) opposed to the state moving forward with this plan. Recent public comments on the HDOA's EPA Application for Emergency Exemption⁶ are over 95% against the use of this biopesticide. We are calling for a halt to this project. The scope, risks, and experimental nature of the state's plan require a detailed, comprehensive **Environmental Impact Statement** documenting the impacts to our native birds, wildlife, environment, and public health.

Who will take responsibility if something goes wrong – the federal government, the State of Hawaii, partners in the multi-agency steering committee *Birds, Not Mosquitoes*, private landowners? Adequate studies and research have not been conducted; and safer, less experimental alternatives have not been considered. Conflicts of interest have not been disclosed or addressed, and the state is rushing forward with this risky, experimental project without the consent of the people of these islands.



Per Article XI of the Constitution of the State of Hawaii¹³ – Conservation, Control and Development of Resources:

CONSERVATION AND DEVELOPMENT OF RESOURCES

Section 1. For the benefit of present and future generations, the State and its political subdivisions shall conserve and protect Hawaii's natural beauty and all natural resources, including land, water, air, minerals and energy sources, and shall promote the development and utilization of these resources in a manner consistent with their conservation and in furtherance of the self-sufficiency of the State.

ENVIRONMENTAL RIGHTS

Section 9. Each person has the right to a clean and healthful environment, as defined by laws relating to environmental quality, including control of pollution and conservation, protection and enhancement of natural resources. Any person may enforce this right against any party, public or private, through appropriate legal proceedings, subject to reasonable limitations and regulation as provided by law.

I do not accept the Anticipated Finding of No Significant Impact (DEA-AFONSI) for the "Suppression of Nonnative Wild Mosquito Populations to Reduce Transmission of Avian Malaria to Threatened and Endangered Forest Birds on East Maui" Environmental Assessment. I demand an Environmental Impact Statement.

Mahalo, Tina Lia Founder & President Hawaii Unites Kihei, Maui, HI

References:

- "Wolbachia infection in wild mosquitoes (Diptera: Culicidae): implications for transmission modes and hostendosymbiont associations in Singapore" – Huicong Ding, Huiqing Yeo, Nalini Puniamoorthy (BMC, 12/09/2020) https://parasitesandvectors.biomedcentral.com/articles/10.1186/s13071-020-04466-8
- "Wolbachia Can Enhance Plasmodium Infection in Mosquitoes: Implications for Malaria Control?" Grant L. Hughes, Ana Rivero, Jason L. Rasgon (PLOS Pathogens, 09/04/2014) <u>https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4154766/</u>
- "Wolbachia-mediated sterility suppresses Aedes aegypti populations in the urban tropics" (preprint) Singapore Consortium, Ng Lee Ching (medRxiv, 06/17/2021) <u>https://www.medrxiv.org/content/10.1101/2021.06.16.21257922v1.full</u>
- "Horizontal gene transfer between Wolbachia and the mosquito Aedes aegypti" Lisa Klasson, Zakaria Kambris, Peter E Cook, Thomas Walker, Steven P Sinkins (BMC Genomics, 01/20/2009) <u>https://bmcgenomics.biomedcentral.com/articles/10.1186/1471-2164-10-33</u>
- "Wolbachia Enhances West Nile Virus (WNV) Infection in the Mosquito Culex tarsalis" Brittany L. Dodson, Grant L. Hughes, Oluwatobi Paul, Amy C. Matacchiero, Laura D. Kramer, Jason L. Rasgon (PLOS Neglected Tropical Diseases, 07/10/2014) <u>https://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0002965</u>



- 6. "DQB Males (Wolbachia pipientis, DQB Strain, Contained in Live Adult Culex quinquefasciatus Males); Receipt of Application for Emergency Exemption, Solicitation of Public Comment" – Environmental Protection Agency (Federal Register, 12/15/22) <u>https://www.federalregister.gov/documents/2022/12/15/2022-27220/dqb-males-wolbachia-pipientis-dqbstrain-contained-in-live-adult-culex-quinquefasciatus-males</u>
- "Wolbachia Horizontal Transmission Events in Ants: What Do We Know and What Can We Learn?" Sarah J. A. Tolley, Peter Nonacs, Panagiotis Sapountzis (Frontiers in Microbiology, 03/06/2019) <u>https://www.frontiersin.org/articles/10.3389/fmicb.2019.00296/full</u>
- "The Intracellular Bacterium Wolbachia Uses Parasitoid Wasps as Phoretic Vectors for Efficient Horizontal Transmission" – Muhammad Z. Ahmed, Shao-Jian Li, Xia Xue, Xiang-Jie Yin, Shun-Xiang Ren, Francis M. Jiggins, Jaco M. Greeff, Bao-Li Qiu (National Center for Biotechnology Information, National Library of Medicine, 02/12/2015) https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4347858/
- 9. "Parasites brainwash grasshoppers into death dive" Shaoni Bhattacharya (New Scientist, 08/31/2005) https://www.newscientist.com/article/dn7927-parasites-brainwash-grasshoppers-into-death-dive/
- "Permit Application for Restricted Commodities into Hawaii" Department of Land and Natural Resources (HDOA, 04/01/22) https://hdoa.hawaii.gov/wp-content/uploads/2022/06/Submittals-6-28-22.pdf
- 11. "Emerging Mosquito Control Technologies" (EPA, 07/19/22) https://www.epa.gov/regulation-biotechnology-under-tsca-and-fifra/emerging-mosquito-control-technologies
- 12. "U.S. Department of the Interior Strategy for Preventing the Extinction of Hawaiian Forest Birds" National Park Service, U.S. Fish and Wildlife Service, U.S. Geological Survey, Office of Native Hawaiian Relations (U.S. Fish and Wildlife Service, 12/15/22) <u>https://www.fws.gov/sites/default/files/documents/DOI%20Strategy%20for%20Preventing%20the%20Extinction n%20of%20Hawaiian%20Forest%20Birds%20%28508%29.pdf</u>
- 13. Constitution of the State of Hawaii, Article XI https://lrb.hawaii.gov/constitution/#articlexi