Integrated Mosquito Management Program

APPENDIX

E

ALTERNATIVES ANALYSIS REPORT

Document Information

Prepared by	Alameda County Mosquito Abatement District
Project Name	Integrated Mosquito Management Program Draft Programmatic Environmental Impact Report
Date	July 2014

Prepared by:

Alameda County Mosquito Abatement District
23187 Connecticut St., Hayward, CA 94545 USA
www.mosquitoes.org

With assistance from

Cardno ENTRIX
2300 Clayton Road, Suite 200, Concord, CA 94520 USA
www.cardno.com

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Acronyms and Abbreviations

ATSB	attractive toxic sugar bait
ATV	all-terrain vehicle
BAAQMD	Bay Area Air Quality Management District
BCDC	San Francisco Bay Conservation and Development Commission
Bs	*Bacillus sphaericus*
Bti	*Bacillus thuringiensis* var. *israelensis*
CDC	Centers for Disease Control and Prevention
CEQA	California Environmental Quality Act
CESA	California Endangered Species Act
CO_2	carbon dioxide
DEET	N,N-diethyl-m-toluamide
District	Napa County Mosquito Abatement District
EC_{50}	median effective concentration
IGR	Insect Growth Regulator
IMMP	Integrated Mosquito Management Program
IPM	Integrated Pest Management
IMM	Integrated Mosquito Management
JH	Juvenile Hormone
LC_{50}	lethal concentration 50 percent
LD_{50}	lethal dose 50 percent
LSAA	Lake and Streambed Alteration Agreement
mg/kg	milligram(s) per kilogram
mg/L	milligram(s) per liter
NOEC	no observable effect concentration
NPDES	National Pollution Discharge Elimination System
OMWM	open marsh water management
PEIR	Programmatic Environmental Impact Report
RIM	rotational impoundment management
SIT	sterile insect technique
ULV	ultra low volume
USACE	U.S. Army Corps of Engineers

Introduction

This report documents the analysis of alternatives for the control of mosquitoes within the Alameda County Mosquito Abatement District's immediate Service Area and, upon request, by agencies in adjacent counties to assist in those areas as well. The Service Area and the adjacent counties are called the Program Area for purposes of environmental impact analysis under the California Environmental Quality Act (CEQA). This report is provided as Appendix E, Alternatives Analysis Report, to the District's Programmatic Environmental Impact Report (PEIR). It presents an evaluation of potential alternatives or "tools," reviews of available literature, and screening criteria to produce recommended components of the Proposed Program. These components represent a reasonable range of alternatives to be discussed in the environmental consequences/impacts sections of the PEIR on the entirety of the District's Program. The conclusions about feasibility of the alternatives and their reasonableness for inclusion in the District's Integrated Mosquito Management Program (IMMP) are those of District staff.

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1 Program Background

The Alameda County Mosquito Abatement District (District) has evaluated a range of control methods for mosquitoes, vectors of human disease and discomfort in its Service Area, located in Alameda County. The District will continue to develop the most effective strategy and methods or “tools” to achieve Program objectives to protect human and animal health.

1.1 Program Location

The District’s Program Area[1] is located in the following counties of the state of California: Alameda, Contra Costa, San Joaquin, Santa Clara, and Stanislaus. The areas proposed for most of the control activities cover 812 square miles in Alameda County. These activities would be focused in the areas with the greatest problems based on monitoring of mosquito populations and testing for presence of the disease pathogens.

1.2 Program History

The District was established in 1930 to provide mosquito control services to the residents and businesses of Alameda County.

The District’s Program is an ongoing series of related actions for control of mosquitoes, vectors of human disease and discomfort. The District’s activities involve the identification of mosquito problems; responsive actions to control existing populations, prevent new sources from developing, and manage habitat to minimize mosquito production; education of landowners and others on measures to minimize mosquito production or interactions; and administration of funding and institutional support necessary to accomplish District objectives.

The District has, and continues to take, an integrated systems approach to mosquito control, using a suite of tools that consists of surveillance, vegetation management, and physical, biological, and chemical controls along with public education. These Program “tools” or components are described in the subsequent subsection as “Program alternatives” for the CEQA process (except for public education, which is exempt from CEQA). Program implementation is weighted heavily towards using those tools that reduce the need for chemical control (e.g., vegetation management, physical and biological control, and public education), thereby minimizing the potential for environmental impacts. To realize effective and environmentally sound mosquito management, mosquito control must be based on several factors:

1. Proactive approach to mosquito management
2. Careful monitoring of mosquito abundance and/or their potential contact with people
3. Continual review and use of treatment criteria (thresholds)
4. Selection of appropriate tools from a wide range of control methods

This ongoing Program consists of a dynamic combination of surveillance, treatment criteria, and use of multiple control activities in a coordinated program with public education that is generally known as Integrated Pest Management (IPM) or Integrated Mosquito Management (IMM).

While these Program components or tools together encompass the District’s IMM Program or IMMP, it is important to acknowledge that the specific tools used by District staff vary from day to day and from site to

[1] The Program Area includes adjacent counties where the District could be requested to provide control services and represents the area of potential environmental impact to be addressed under the California Environmental Quality Act (CEQA).

site in response to the mosquito species that are active, their population size or density, their age structure, location, time of year, local climate and weather, potential for mosquito-borne disease, proximity to human populations, including (a) proximity to sensitive receptors, (b) access by District staff to mosquito habitat, (c) abundance of natural predators, (d) availability and cost of control methods, (e) effectiveness of previous control efforts at the site, (f) potential for development of resistance in mosquito populations, (g) landowner policies or concerns, (h) proximity to special status species, and (i) applicability of Endangered Species Recovery Plans, Habitat Conservation Plans, Natural Community Conservation Plans, and local community concerns, among other variables. Therefore, the specific actions taken in response to current or potential mosquito activity at a specific place and time depend on factors of mosquito and pathogen biology, physical and biotic environment, human settlement patterns, local standards, available control methods, and institutional and legal constraints. While some consistent mosquito sources are exposed to repeated control activity, many areas with minor mosquito activity are not routinely treated, and most of the land within the District's Service Area has never been directly treated for mosquitoes.

Also, note that an essential component of an effective and environmentally sensitive IMMP is the District's proactive approach to the management of mosquito populations. Such an approach requires continuous surveillance, early detection, a rapid response, and a good public education program to minimize human and mosquito interactions. Most importantly, a proactive approach minimizes the use of pesticides while maximizing the opportunities to use those methodologies that are the least disruptive to nontarget organisms and the environment.

2 Potential Tools

Potential tools for use in the Program are described below and include measures used for other similar control programs in California. This chapter presents a description of each tool. The evaluation of each as to whether it is applicable to or an effective component of a mosquito control program is presented here and explained further in Section 3.

2.1 Integrated Pest Management

2.1.1 Description

Integrated pest management (IPM) is a decision making process that emphasizes an ecosystem approach to maintain the population of any pest at or below the level that causes damage while also minimizing societal and environmental impacts. The focus is not on how to kill the pest. Instead, an ecosystem analysis is used that looks at all factors in a pest's environment that allow it to thrive (i.e., food resources, water, habitat) as well as those factors that compete with the pest (i.e., predators, parasites, diseases). IPM recognizes that if one component of an ecosystem is changed other parts of the ecosystem will be affected. Therefore, in order to achieve effective long-term pest management results and avoid unintended negative side effects of pest treatment to humans and environmental resources, the pest itself, its relationship to other organisms in the ecosystem, and the factors in its environment that allow it to survive and reproduce must be considered.

IPM uses one or more tools to prevent pest numbers from reaching damaging levels. The keys to a successful IPM program are: selecting a proactive approach; identifying the pest; understanding pest biology, behavior, and population dynamics; monitoring pest numbers; establishing a treatment threshold level to trigger actions that will prevent damage or loss; selecting the appropriate tool to prevent pest numbers from reaching harmful levels; implementing management tools in a timely manner; following-up with evaluation on effectiveness and any unintended impacts of management actions taken; and recognizing that often some damage due to pest presence may occur and is acceptable.

2.1.2 Examples of Tool Use

IPM is used by all mosquito and vector control agencies in California.

2.1.3 Applicability to District IMMP

The District's current IMMP uses key IPM concepts. Where IPM controls pest numbers and believes that some damage is acceptable, the District has instances in which the threat to public health requires additional measures beyond traditional IPM (e.g. impacts of a natural disaster such as a devastating earthquake or a weather event that results in flooding, declaration of a public health emergency by state health officials).

2.2 Mosquito Surveillance

2.2.1 Description

Mosquito surveillance, which is an integral part of the District's responsibility to protect public health and welfare, involves monitoring mosquito populations, their habitat and disease pathogens, and human-mosquito interactions. Mosquito surveillance provides the District with valuable information on what mosquito species are present or likely to occur, when they occur, where they occur, how many they are, and if they are carrying disease or otherwise affecting humans. Mosquito surveillance is critical to an IMM program because the information it provides is evaluated against treatment criteria to decide when and where to institute mosquito control measures. Information gained is used to help form action plans that can also assist in reducing the risk of contracting disease. Equally important is the use of mosquito surveillance in evaluating the efficacy, cost effectiveness, and environmental impacts of specific mosquito control actions.

2.2.2 Examples of Tool Use

Examples include field counting/sampling and trapping, arbovirus surveillance, field inspection of known or suspected habitats, and public service requests.

2.2.3 Applicability to District IMMP

Already used under current Program.

2.3 Physical Control

2.3.1 Description

Physical control is managing mosquito habitat to reduce mosquito production or migration through “source control’ measures that are nonchemical or nonbiological techniques. In many cases, physical control activities involve restoration and enhancement of natural ecological functioning. This tool is often the most cost-effective and least toxic element of an integrated mosquito management program. Physical control activities can be generally divided into water management, exclusionary practices and physical barriers, and management of man-made containers.

2.3.1.1 Water Management/Control Structures

Water management techniques are used primarily for the control of mosquito populations and include, but are not limited to, water control and maintenance of channels, tide gates, levees, and other water control facilities to improve water circulation. In natural settings, these activities are generally used to reduce mosquito production while at the same time improving habitat values for many predators and parasites of larval mosquitoes. In artificial or highly managed settings (e.g., dredge disposal ponds, diked marshes, duck clubs), physical control can include improved drainage as well, to reduce the duration of standing water below the time needed for the development of immature mosquitoes.

Water management is a core component of effective mosquito control. For nondomestic sources, this part of the District's IMMP involves participation in the planning, implementation, and maintenance of stormwater detention facilities, riparian corridors, and managed and tidal wetlands. The District provides information on design and maintenance strategies that minimize the potential for creation of mosquito habitat while optimizing support for the desired function(s) of the water structure, feature, or habitat being restored or created. In some instances, the District may also partner with resource managers (i.e., federal, state, and local agencies) and contribute additional resources, such as labor and materials, towards those projects that enhance or create habitats for wildlife as well as public benefit. These types of projects usually involve wetlands such as tidal marshes, freshwater marshes, and riparian corridors and may include activities such as creation of impoundments, installation of water control structures, ditching, brushing, and removal of nonnative plants, and planting of native vegetation.

Restoration or enhancement of hydrological functioning of wetlands by means of ditching and/or removal of old ditches constitutes the majority of the District's physical water management activities. Ditching can be accomplished by hand or with the use of a speed scavel or rotary ditcher and may also involve open marsh water management (OMWM) or rotational impoundment management (RIM) principles and techniques. Grid or parallel ditching, a technique that was used many decades ago by some US mosquito control agencies, has not been used by the District as an ecosystem approach to water management rather than simply dewatering a mosquito-breeding site has and continues to be emphasized. Thus, the District carefully places mosquito management ditches, taking into account many factors including, but not limited to, size, depth, width, natural curves or sigmoidal nature, generated spoils, tidal flow, location of desired ponding areas, impacts to native flora and fauna, and potential for access by invasive or nonnative species.

Numerous benefits are associated with carefully managing water circulation and levels within natural and some man-made mosquito habitats, the four most prominent being the significant reduction of pesticide use that would otherwise be required to minimize human-mosquito interactions, significant long-term reduction of mosquito population levels, reduced potential for disturbance, and the enhancement of wetland functions. Evaluation of physical control methods such as hand and rotary ditching as well as OMWM and RIM indicates that they have minimal significant detrimental impact of the environment when performed under District and permitting agency guidelines and, on the contrary, are generally beneficial to a wide range of desirable species including special status species (Balling et al. 1979, 1980; Balling and Resh 1982, 1983, 1991; Barnby and Resh 1980; Barnby et al. 1985; Batzer and Resh 1992; Batzer et al. 1993, 1997; Collins and Resh 1985; Collins et al. 1986; Kramer et al. 1992, 1995; Resh and Balling 1979, 1983a,b; Resh et al. 1980). Wolfe (1996) reviews the effects of OMWM on tidal marsh resources and includes in his discussion data and comparisons with grid ditching, runneling (a form of hand ditching), and modified versions of OMWM. He concludes that OMWM is an effective tool for long-term management of salt marsh mosquitoes with no significant negative impacts to other wetland resources. He also points out that the use of OMWM must be customized to meet the specific conditions of the marsh type and management need as habitat heterogeneity within a marsh, and the differences between marsh types, precludes a one-size-fits-all approach to water management for mosquitoes in tidal and managed wetland systems.

Tonjes (2013) extensively reviews the literature discussing the impacts ditching has had on salt marshes of the eastern US. He points out the complexity of biotic and abiotic variables that affect interpretation and understanding of available data and states that the qualitative generalizations made from published studies suggests that ditching does affect certain marsh elements. Changes to water table height as a result of ditch placement, design, depth and number can result in short-term, and sometimes long-term, changes to vegetation as well as composition and distribution of avian, fish, and invertebrate populations. Obligate marsh species and those organisms dependent on certain marsh features are more sensitive and can be both negatively and positively impacted. Potential marsh acidification, changes in soil salinity, carbon export, and water quality issues (e.g., releases of reduced forms of nitrogen and stored pollutants in marsh sediments) are other areas for research as variable data demonstrate both negative and positive potential impacts. Lastly, some effects are not always as clear as they may seem or as directly linked to a particular cause or action. Therefore, careful planning, coordination, and adaptive management are essential to effectively manage the marsh ecosystem as a whole rather than for one or a few special status and mosquito species. An ecosystem approach is a core tenet of the District's IMMP and every effort is made to protect natural resources while at the same time working with mosquito populations to minimize human-mosquito interactions and potential health issues.

Careful water management in tidal, managed, and some riparian habitats for mosquito control typically provides long-term effective control but still requires continual surveillance and occasional maintenance for continued effectiveness. This type of work typically requires permitting and oversight by various state and federal regulatory agencies that help the District to address potential habitat, sensitive species, and water quality issues. This coordinated effort allows the District to meet its goal of being an effective resource manager. The District recognizes that wetland systems are highly complex and no single management strategy is without potential detrimental and beneficial impacts.

2.3.1.2 Exclusion/Physical Barriers

Another form of physical control is the practice of exclusion or creating physical barriers that prevent mosquitoes from gaining access to water sources or from feeding on people and their pets. This control can involve the use of screens (e.g., door, windows, septic vents) and sealing off other points of entry (e.g., around pipes, drains). The District performs inspections of properties and provides information to assist property owners with the exclusion techniques but does not, however, perform this type of work.

2.3.1.3 Management of Man-Made Containers

This aspect of physical control involves the proper draining and/or removal of containers (e.g., buckets, barrels, ornamental ponds, fountains, wading pools, spas, tires, abandoned appliances) that hold water capable of producing mosquitoes. Sometimes containers holding water can simply be overturned, other times holes are drilled to facilitate drainage prior to removal and disposal. The benefits of this activity go far beyond just managing mosquito populations as it also includes educating landowners about the mosquito issues associated with improperly maintained or discarded materials. Management of man-made containers is the most frequent peridomestic mosquito control technique used by the District.

2.3.2 Examples of Tool Use

Physical control includes coordination with landowners, managers, and regulatory authorities for repair of weir structures in tidal and seasonal wetlands managed for waterfowl; repair of culverts and tide gates in tidal wetlands; hand removal of sediment buildup in small ditches and channels in tidal marshes and agricultural ditches; and the removal of obstructive debris such as boards, tires, small containers, and other man-made objects that can obstruct proper water flow and/or interfere with the functioning of water control structures. Large mechanized equipment (e.g., rotary ditcher and low ground pressure caterpillar tractor) for the maintenance of ditches, channels, and levees has not been used in more than 20 years but is a tool that can be used as part of physical control activities. Therefore, heavy mechanized equipment is a tool for potential future use. Recommended exclusionary practices include, but are not limited to, screening windows, doors, and water barrels/containers and installing fans over doorways and windows

2.3.3 Applicability to District IMMP

Physical control is an essential part of the District's IMMP and is also an important part of the District's public education program. This tool can involve the District performing the actual work and/or making recommendations to landowners, land managers, and resource agencies concerning mosquito habitat management.

Physical manipulation of wetlands requires careful planning, coordination with landowners and regulatory agencies, the appropriate permits, and timely and proper execution of the work to assure preservation of wetland functions and resources. Actions that benefit one or a few species have the potential to negatively affect others and/or may also result in shifts over time to certain wetland attributes. Therefore, the District consults with landowners/managers and all appropriate resource agencies to make sure that sensitive species and their habitats have been identified and that all appropriate permits are in place prior to commencement of work. Additionally, any recommendations made by the District to landowners and land managers concerning physical control activities also include the need for their consultation with regulatory agencies and the acquisition of any potential permits that may be required.

Under some circumstances, physical control techniques are not practical or can become ineffective. For example, large breeding sites such as seasonal wetlands, marshes, stock ponds and wastewater ponds cannot be screened to prevent mosquito access and breeding. The use of screens is effective for stopping mosquitoes from accessing small potential breeding sites such as rainwater barrels and septic tanks. Screening also prevents mosquitoes from gaining entry to the home and workplace but does not protect people or their pets when they are outside. Physical exclusion is very dependent on good public outreach, people being disciplined about maintaining screens in good working condition and keeping them in place, continually being vigilant about the condition of potential points of entry for mosquitoes, and taking timely appropriate action.

The District's diligent public outreach helps effectively manage peridomestic habitats that can produce unwanted mosquitoes. Routine consultation with various biologists and wetlands experts helps the District ascertain the best methods for meeting its goal of being a good steward of wetland resources while also managing mosquito populations that may be present within different wetland habitats.

2.4 Vegetation Management

2.4.1 Description

The species composition and density of vegetation are basic elements of the habitat value of any area for mosquitoes, for predators of mosquitoes, and for protected flora and fauna. District staff periodically undertakes vegetation management activities as a tool to reduce the habitat value of sites for mosquitoes or to aid production or dispersal of mosquito predators, as well as to allow access by District staff to mosquito habitat for surveillance and other control activities. Vegetation management can be accomplished by using hand tools (mechanical removal), herbicides, or burning.

2.4.1.1 Mechanical

Mechanical vegetation management by District staff generally consists of activities to reduce the mosquito habitat value of sites by improving water circulation or access by fish and other predators, or to allow access by District staff to standing water for inspections and treatment. For vegetation management, the District uses hand tools or other mechanical means (i.e., heavy equipment) for vegetation removal or thinning.

2.4.1.2 Herbicides

Herbicides (chemical pesticides with specific toxicity to plants) may be used to help reduce the density of emergent vegetation in aquatic habitats (e.g., winery waste ponds, dairy lagoons) that provide harborage for mosquitoes and prevent the effective use of biological control agents or mosquito larvicides. Herbicides may also be used to manage vegetative growth that prevents adequate and fire safe access to mosquito-breeding habitats. The use of herbicides for the purpose of habitat access has not been used in the past but may be occasionally used in the future and, therefore, remains a part of the District's IMMP.

An Ecological & Human Health Assessment Report was prepared by Cardno ENTRIX as Appendix B to the District's upcoming PEIR. This report (in Section 4.6) evaluated the potential hazards and estimated risks of 13 herbicide active ingredients, including glyphosate and imazapyr. Documented toxicity and environmental fate of these herbicides were reviewed and evaluated. Those that demonstrated reasonable efficacy with minimal undesirable effects (i.e., unwanted estimated risk) were selected for inclusion in the District's IMMP.

The District does not use pre-emergent herbicides (e.g., diuron) as a part of its IMMP. Post-emergent herbicides (e.g., glyphosate and imazapyr) may be used but are a least preferred method for the management of vegetation in mosquito habitats. When post-emergent herbicides are used, their application usually occurs in aquatic habitats such as winery waste ponds and other types of isolated man-made terminal waterbodies.

2.4.1.3 Burning

Burning, though rarely planned on being used, is another means by which the District may thin out vegetation in aquatic habitats. This technique is useful for large ditches and man-made ponds that are completely choked with very dense stands of cattails and weeds. This strategy of vegetation management would be carefully coordinated with local fire departments, the landowner, and the Bay Area Air Quality Management District prior to implementation. Burning takes place usually during the late fall or early winter.

2.4.2 Examples of Tool Use

Vegetation management includes coordination with landowners, managers, and regulatory authorities for trimming vegetation with gas-powered weed trimmers, hand saws, small chain saws, and hand pruners to remove vegetative growth obstructing channels and ditches and access to potential mosquito-breeding sites as well as reducing the quality of mosquito habitat; the application of herbicides to manage emergent weed growth from wastewater ponds; the application of herbicides on unpaved access roads to facilitate access to mosquito breeding sources and minimize the risk of fires started by hot vehicles traveling through tall dead weeds; and burning of dense stands of cattails and weeds in large ditches and man-made ponds.

2.4.3 Applicability to District IMMP

Vegetation management, usually requires a combination of mechanical removal, application of herbicides, and occasional burning to be effective at allowing predator access to immature mosquitoes that use dense emergent vegetation for protection. No one technique is long lasting, nor does long-term effectiveness occur when they are used in combination. Extraordinarily dense stands of emergent vegetation are either mechanically removed or burned (depending on the size and access of the site to equipment, safety concerns, and the presence of sensitive habitats and species) and then managed on an annual or biennial basis using mechanical removal, herbicides, or both as needed. The District prefers to make recommendations to landowners and managers rather than using this control strategy directly and encourages the use of water management practices that discourage excessive growth of emergent vegetation and weeds. The redesign or restoration of wetland sites that enhances their functioning and species diversity while minimizing mosquito habitat is a core tenet of the District's IMMP. Studies in constructed treatment wetlands have shown that these systems are complex and that no one vegetation management technique provides long-term effective mosquito control without including water management and design strategies that incorporate species of vegetation used, water depth, ratio of vegetative cover to open water areas, steepness of wetland margins and bottom slopes, wetland type, size, etc. (Jiannino and Walton 2004; Knight et al. 2003; Russell 1999; Thullen et al. 2002; Walton et al. 2012; Walton and Workman 1998).

Additionally, the District recognizes that constructed, restored, and natural wetlands are significant to a wide range of fauna such as waterfowl, amphibians, various other invertebrates (e.g., insects, crustaceans), and fish. Also, a difference in the use as well as seasonality of use occurs with different types of constructed and natural wetlands, and water quality, hydroperiod, types of vegetation, water depth, and percent open water versus vegetative cover play a significant role to the fauna that use these habitats (Batzer and Resh 1994; Melvin and Webb 1998; Reaves and Croteau-Hartman 1994).

The use of herbicides is one of three integrated components used for the management of vegetation associated with access to and within mosquito habitats. The restricted use of these materials to primarily man-made sites precludes the risk of damage to, or loss of, sensitive wetland habitats or special status species. The one exception is for the management of invasive, nonnative weeds (e.g., spartina, pepperweed, arundo) and is coordinated with and overseen by regulatory or other governmental entities (e.g., California Department of Fish and Wildlife, US Fish and Wildlife Service, Flood Control Districts, and Park Districts). Surveys are conducted to ensure that application of these chemicals will not risk impacting any sensitive habitats, special status species, or food crops. Furthermore, care is taken by District personnel and contractors to make sure that potential drift is managed by using these chemicals only during periods with little or no wind (less than 5 miles per hour maximum). Application is also timed to maximize effectiveness and reduce the need for additional applications in the same area.

Depending on location, habitat, and potential for presence of special status species, vegetation management may require permits and agreements (e.g., USACE, CESA, BCDC, NPDES, LSAA, BAAQMD) prior to commencement of work. The District consults with landowners/managers and all appropriate resource agencies to make sure that sensitive species and their habitats have been identified and that all appropriate permits are in place. Any recommendations made to landowners and land managers by the District also include their need for consultation with regulatory agencies and the acquisition of any potential permits that may be required.

2.5 Biological Control Pathogens (Viruses)

2.5.1 Description

Mosquito viral pathogens are highly host-specific and usually infect mosquito larvae when they are ingested. Upon entering the host, these pathogens multiply rapidly, destroying internal organs and consuming nutrients. The pathogen can be spread to other mosquito larvae in some cases when larval tissue disintegrates and the pathogens are released into the water to be ingested by uninfected larvae. Becnel and White (2007) provide a thorough summary of the current understanding and last 20 years of research concerning mosquito pathogenic viruses. Their review indicates numerous issues still need addressing before mosquito viral pathogens can be used as an effective mosquito control strategy.

2.5.2 Examples of Tool Use

Examples of viruses that can infect mosquitoes are mosquito iridoviruses, densonucleosis viruses, nuclear polyhedrosis viruses, cytoplasmic polyhedrosis viruses, and entomopoxviruses.

2.5.3 Applicability to District IMMP

Pathogenic viruses are not commercially available for mosquito control at present. Therefore, viral pathogens are not a part of the District's IMMP at this time.

2.6 Biological Control Pathogens (Bacteria)

2.6.1 Description

Bacterial mosquito pathogens are generally host-specific and usually infect mosquito larvae when they are ingested. Upon entering the host, these pathogens multiply rapidly, destroying internal organs and consuming nutrients. The pathogen can be spread to other mosquito larvae in some cases when larval tissue disintegrates and the pathogens are released into the water to be ingested by uninfected larvae. Environmental factors such as salinity, low temperatures, high larval densities, life stage (age) of the mosquito, and dense vegetative cover that interferes with application at the mosquito-breeding site can limit the effectiveness and presence of certain bacterial pathogens of mosquitoes. For example, *Bacillus sphaericus* (Bs) works best in highly polluted waters but not very well in brackish or saline environments. The species of mosquito may also play a role in effectiveness (e.g., several species of *Aedes* mosquitoes, including salt marsh *Aedes*, are not very susceptible to the larvicide Bs) (Baumann et al. 1991; Brown et al. 2004; Davidson 1989; Lacey et al. 1988; Mittal 2003).

2.6.1.1 Bacillus sphaericus (Bs)

Bs is a commonly occurring spore-forming bacterium found throughout the world in soil and aquatic environments. Certain strains of this bacterium produce a protein endotoxin, which is pathogenic to immature mosquitoes. This endotoxin destroys the insect's gut in a way similar to the protein crystals of *Bacillus thuringiensis* var. *israelensis* (Bti). That is, the toxin is only active against feeding mosquito larval stages and it must be partially digested before it becomes activated.

Bs adversely affects larval mosquitoes but, in contrast to Bti, is virtually nontoxic to black flies (*Simuliidae*). *Culex* species are the most sensitive to Bs, followed by *Anopheles* and some *Aedes* species. In California, *Culex* spp and *Anopheles* spp may be effectively controlled. Several species of *Aedes* have shown little or no susceptibility, and salt marsh *Aedes* are not susceptible. Bs differs from Bti in being able to control mosquito larvae in highly organic aquatic environments, including sewage waste lagoons, animal waste ponds, and septic ditches. Also in contrast to Bti, field evaluations of commercial Bs products (VectoLex) have shown environmental persistence for 2 to 4 weeks, and the ability to recycle (grow and reproduce). This persistence varies with a number of environmental parameters, and is low in saline or highly organic environments. Bs has been extensively tested and has had no adverse effects on mammals or other nontarget organisms (Ali and Nayar 1986; Aly and Mulla 1987; Aly et al. 1985; Holck and Meek 1987; Karch et al. 1990; Key and Scott 1992; Lacey and Merritt 2003; Merritt et al, 2005; Miura et al. 1981; Mulla et al. 1984a,b; Rodcharoen et al. 1991; Shadduck et al. 1980; Siegel and Shadduck 1990; Tietze et al. 1993; Walton and Mulla 1991; Yousten et al. 1991). No mortalities, pathogenicity or treatment-related evidence of toxicological effects were observed in rats administered oral, intravenous or intratracheal doses of technical Bs. The acute oral and dermal lethal dose 50 percent (LD_{50}) values are greater than 5,000 and 2,000 milligrams per kilogram (mg/kg), respectively. Oral exposure of Bs is practically nontoxic to mallard ducks. No mortalities or signs of toxicity occurred following a 9,000-mg/kg oral treatment. Birds fed diets containing 20 percent wet weight of the technical material experienced no apparent pathogenic or toxic effects during a 30-day treatment period. Mallards given an intraperitoneal injection of Bs demonstrated toxicological effects including hypoactivity, tremors, ataxia, and emaciation. The LD_{50} value was greater than 1.5 mg/kg.

Acute aquatic freshwater organism toxicity tests were conducted on bluegill sunfish, rainbow trout, and daphnids. The 96-hour lethal concentration 50 percent (LC_{50}) and no observable effect concentration (NOEC) value for bluegill sunfish and rainbow trout was greater than 15.5 milligrams per liter (mg/L); the 48-hour median effective concentration (EC_{50}) and NOEC value for daphnids was greater than 15.5 mg/L. Acute aquatic saltwater organism toxicity tests were conducted on sheepshead minnows, shrimp, and oysters. The 96-hour LC_{50} value for both sheepshead minnows and shrimp was 71 mg/L, while the NOEC value was 22 mg/L for sheepshead minnows and 50 mg/L for shrimp. The 96-hour EC_{50} value for oysters was 42 mg/L with a NOEC of 15 mg/L. The LC_{50} and NOEC value for immature mayflies was 15.5 mg/L. Honeybees exposed to 10^{-4} to 10^{-8} spores /mL for up to 28 days demonstrated no significant decrease in survival when compared to controls. Additional studies on various microorganisms and invertebrates, specifically cladocerans, copepods, ostracods, mayflies, chironomid midges, water beetles, backswimmers, water boatmen, giant water bugs, and crawfish, have shown no adverse effects or negative impacts. Furthermore, Ali (1991) states that although Bs is known to be highly toxic to mosquito larvae, Bs does not offer any potential for midge control. Acute toxicity of Bs to nontarget plants was also evaluated in green algae. The 120-hour EC_{50} and NOEC values exceeded 212 mg/L.

Lacey (2007) reviews the prior 20 years of research concerning Bs toxins, their modes of action and factors affecting activity, resistance, safety, and the role of this entomopathogen in integrated mosquito management programs. He concludes that in many situations this bacterial biological control agent of mosquitoes is an effective alternative to many other broad spectrum mosquito larvicides as it has numerous environmental benefits including safety to nontarget organisms, reduction of pesticide residues, no effect on the activity of other mosquito predators and pathogens, and little or no overall environmental impact. The compatibility of this insecticide with other biological control agents also allows for a more sustainable integrated management approach and it is the cumulative effect of the aforementioned advantages that help to offset the significant increases in costs associated with the use of this insecticide. The one concern is resistance, especially since the Bs toxin apparently binds to a single receptor on the microvilli of the larval midgut. Resistance has been reported in a number of regions throughout the world (Adak et al. 1995; Chevillon et al. 2001; Mulla et al. 2003; Nielsen-Leroux et al. 2002; Oliveira et al. 2003, 2004; Rao et al. 1995; Silva-Filha et al. 1995; Su and Mulla 2004; Wirth et al. 2000). Therefore, this material must be used with care and routinely rotated with the use of other available insecticides (e.g., Bti) and management strategies to minimize the risk of resistance (Regis and Nielsen-LeRoux 2000; Zahiri et al. 2002).

2.6.2 Examples of Tool Use

The only live bacteria commercially available and pathogenic to mosquitoes is Bs. This material is currently available as a granule (VectoLex CG), water dispersible granule (VectoLex WDG), and water-soluble packet (VectoLex WSP) for the treatment of immature mosquitoes.

2.6.3 Applicability to District IMMP

Bs is a significant and effective immature mosquito management tool. Various formulations of Bs are used as a part of the District's IMMP.

2.7 Biological Control Parasites/Parasitoids

2.7.1 Description

A parasite is an organism that lives in or on another organism (the host) on which it depends for nutrients and survival. Parasites typically harm the host with damage ranging from discomfort to death. Their size is typically smaller and their rate of reproduction is usually much faster than their host. The several types of parasite are loosely defined as follows: (1) Ectoparasite - a parasite that lives on the surface of the host (e.g., mites, lice, and ticks); (2) Endoparasite - a parasite that lives inside the body of the host either within the spaces of the host's body (an intercellular parasite such as tapeworms and nematodes such as heartworms) or within the cells of the hosts body (bacteria and viruses); (3) Epiparasite - a parasite that feeds on another parasite; and (4) Parasitoid - an organism that although initially parasitic on its host, ultimately sterilizes or kills and sometimes consumes its host (e.g., microbial pathogens of insects such as microsporidians and entomophthora fungus).

2.7.1.1 Mosquito Parasites

The life cycles of mosquito parasites are biologically more complex than those of mosquito pathogens and involve intermediate hosts, organisms other than mosquitoes. Mosquito parasites are ingested by the feeding larva or actively penetrate the larval cuticle to gain access to the host interior. Once inside the host, parasites consume the internal organs and food reserves until the parasite's developmental process is complete. The host is killed when the parasite reaches maturity and leaves the host (*Romanomermis culicivorax*) or reproduces (*Lagenidium giganteum*). Once free of the host, the parasite can remain dormant in the environment until it can begin its developmental cycle in another host.

2.7.1.2 Lagenidium giganteum

Lagenidium giganteum, an oomycete fungus, was briefly available under the trade name Laginex. Production, storage (shelf life), registration, and costs were some of the issues limiting the use of this parasite for mosquito control. Other factors included the environmental limitations of temperature (less than 16 or more than 32 degrees Celsius), moderate salinity levels (less than 10 parts per thousand), and moderate organic content of the water (Kerwin 2007; Merriam and Axtell 1982). Scholte et al. (2004) reviews the different entomopathogenic fungi that have been studied for mosquito control purposes and states an ideal fungus for mosquito control has nine key features. They are (1) kills adult and larval stages; (2) requires no more than a few applications per season; (3) is easily dispersed by adult female mosquitoes to uninfected breeding sites; (4) shows residual activity and persistence in mosquito populations after introduction; (5) kills only mosquitoes; (6) is effective over a wide range of salinity, temperature, humidity, and water quality conditions; (7) is easily and cost-effectively mass produced; (8) has a long shelf-life and can be easily stored; and (9) is not harmful to humans or other nontarget organisms. Scholte concludes by stating that "none of the mosquito-pathogenic fungi presently known exhibit all of these characteristics, but they all exhibit at least some."

2.7.1.3 Other Fungi

Other fungi, including the recently reclassified microsporidia, continue to be found and studied for their potential as biological control agents. Andreadis (2007) and Scholte et al. (2004) provide thorough updated reviews of the entomopathogenic fungi of mosquitoes. Elucidation of their complex life histories and effectiveness as biological control agents of mosquitoes (e.g., *Coelomomyces* spp, *Culicinomyces* spp, and certain microsporidia) are discussed. As mentioned above, some technical issues still need solving before these biological control agents can be commercially produced and available for use.

2.7.1.4 Lambornella clarkii

Lambornella clarki, a protozoan, has been studied as a biological control agent of container-breeding mosquitoes, especially the western treehole mosquito, a natural host of this endoparasitic ciliate (Washburn and Anderson 1986, 1990a,b; Washburn et al. 1988). This parasite has cysts that are resistant to desiccation and, therefore, allow this ciliate to persist to the next year. Production and storage methods investigations and early field trials have been conducted to determine the efficacy of this ciliate for biological control (Anderson et al. 1986a,b, 1989; Anderson and Washburn 1989a,b, 1990). Although the data demonstrate that *L. clarki* appears to be a promising biological control agent, it is at this time not commercially available for use.

2.7.1.5 Nematodes

Mermithid nematodes, especially *Romanomermis* spp and *Reesimermis* spp, have received a fair amount of study for use as biological control agents of mosquitoes, with *Romanomermis culicivorax* commercially produced as Skeeter Doom for a short time many years ago. Although this nematode showed much promise, the following limitations still restrict its widespread use: low salinity levels, organically rich waters with low oxygen levels, predation by other aquatic organisms, the potential for the development of host resistance, and the costs associated with mass in-vivo production (Legner 1995; Petersen 1978; Petersen and Willis 1970; Platzer 1981, 2007).

2.7.2 <u>Examples of Tool Use</u>

Examples of mosquito parasites are the fungi *Lagenidium giganteum, Coelomomyces* spp*, Culicinomyces clavosporus*, *Metarhizium anisopliae*, *Pythium* spp, *Tolypocladium cylindrosporum*, *Nosema algerae, Hazardia milleh, Vavraia culicis, Amblyospora californica, Edhazardia aedis,* and *Parathelohania* spp*;* the protozoa *Lambornella clarki*, *Sarcosystis singaporensis* and *Tetrahymena* spp; the nematodes *Capillaria hepatica*, *Reesimermis nielseni* and *Romanomermis culicivorax;* and the wasp *Sphecophaga vesparum vesparum*.

2.7.3 <u>Applicability to District IMMP</u>

Many parasites of mosquitoes exist, some whose biologies are still not fully understood. Research in this area, especially for mosquito control, has been and continues to be significant as additional environmentally friendly tools are sought to help manage mosquito populations. Although the use of parasites as a means for managing mosquito populations shows promise, much work concerning their biology, cultivation, mass production, transport, and release remains to be done.

Parasites are not readily available commercially for mosquito or yellow jacket control at present. Therefore, they are not a part of the District's IMMP at this time.

2.8 Biological Control Predators

2.8.1 <u>Description</u>

Mosquito predators are represented by highly complex organisms, such as insects, fish, birds, and bats that may consume immature or adult mosquitoes as prey. Predators are opportunistic in their feeding habits and typically forage on a variety of prey types, which allows them to build and maintain populations at levels sufficient to help control mosquitoes, even when mosquitoes are scarce. The ability of predators to effectively control Mosquito organisms is related to four factors: (1) whether mosquitoes are preferred prey, (2) whether the hunting strategy of the predator maximizes contact with mosquitoes, (3) whether the predator consumes large numbers of mosquitoes, and (4) whether the predator is present in sufficient numbers to control mosquitoes. Predator effectiveness is enhanced when proper conditions are present.

The overall objective of using predators is to reduce the frequency of pesticide applications as well as human-mosquito interactions and associated health issues, which minimizes the risk of potential environmental impacts associated with some pesticides and delays development of mosquito resistance to pesticides. The District recognizes the value of maintaining and promoting as many native predatory species as possible, and this principle is a core component of its IMMP.

Predation on mosquitoes is a natural process that will occur without human intervention. However, the level of mosquito control by natural predators can be increased by the conservation of predators in the environment and by augmentation of the predator population through stocking and habitat enhancement.

2.8.1.1 Mosquito Predators

References to some of the predators of mosquitoes can be found dating back more than 100 years and help form the basis for much of the research that has occurred since (Beutenmuller 1890; Cattell 1903; Felt 1904; Howard 1901, 1910: Mitchell 1907; Smith 1904; Weeks 1890). Therefore, predators as discussed in this section will be loosely grouped into five general categories: invertebrates, amphibians, fish, bats, and birds.

2.8.1.1.1 Invertebrate Predators

Invertebrate predators are the most numerous and commonly encountered predators of mosquitoes within any mosquito-breeding habitat. Their members include, but are not limited to, coelenterates; platyhelminths (flatworms); cyclopoid copepods; insects of many orders, especially Odonata (dragonflies and damselflies), Ephemeroptera (mayflies), Hemiptera (true bugs), Coleoptera (beetles); and spiders. A resurgence of research and evaluation on the various invertebrate predators of immature mosquitoes began in the late 1960s and has continued to this day (Ali and Mulla 1983; Bay 1967, 1969; Collins and Washino 1978; Ellis and Borden 1970; Garcia and Schlinger 1970; Garcia et al. 1974; Hazelrigg 1974; Hokama and Washino 1966; Lee 1967; Legner and Medved 1970, 1974; Miura et al. 1978; Qureshi and Bay 1969; Rayah 1975; Roberts et al. 1967; Sjogren and Legner 1974; Stewart and Miura 1978; Veneski and Washino 1970; Washino 1969a,b; Yu and Legner 1975; Yu et al. 1974; Zalom et al. 1978). Factors such as effectiveness, culture techniques, and release, as well as potential environmental limitations of salinity, temperature, pH, presence/lack of vegetation, substrates, seasonality of flooding and drying of habitats, persistence, and prey selectivity have been examined for a number of mosquito predators.

Quiroz-Martinez and Rodriguez-Castro (2007) provide a nice summary of the use of aquatic insects as predators of immature mosquitoes. Their discussion includes a review of the factors significant to the success of biological control programs for mosquitoes, especially with respect to the predator-prey relationship. These factors are (1) prey preference of the predator, (2) species diversity within the mosquito habitat, (3) aquatic ecosystem stability, (4) density of the larval mosquito population, (5) where the predator typically spends most of its time within the water column, (6) number of predators needed for release and optimal efficacy, (7) recovery of the immature mosquito population, (8) predator-prey synchronization, (9) refugia for both the predator and especially the prey, (10) coevolution of the antipredation responses of the prey and the attack processes of the predator, and (11) community participation in both the planning and operational efforts to help the community understand the roles various biocontrol organisms play in controlling mosquitoes.

Mogi (2007) elaborates further on some of the challenges associated with the use of invertebrate predators. First, is the issue of mass production, storage, and release, especially since most predators are cannibalistic and also require live organisms as a food source. Second, many are opportunistic in their predatory habits and consume a wide variety of prey other than mosquitoes, which can be helpful as it allows predators to survive when mosquito populations are low or altogether absent. Conversely, it can be a disadvantage because predators may not effectively reduce mosquitoes due to the availability of alternative prey, some of which include populations of other beneficial mosquito predators. Third, is the presence of other indigenous predators and the complexity of the relationships and differences in vulnerabilities that exist not only between predator-prey but also amongst the different predator populations within a given ecosystem. Anti-predator behaviors, chemical cues, and other types of interference are not fully understood; and more research is required to better understand what factors contribute to predator effectiveness.

Marten and Reid (2007) provide a good review of the research concerning cyclopoid copepod biology, mass production, storage, field application, and environmental and health impacts. It is important to note that copepods cannot be used in all habitats and, therefore, must be matched to the appropriate mosquito-breeding site to be effective. Equally important, cyclopoid copepods effectively reduce *Aedes* mosquitoes, but are less effective at reducing *Anopheles* spp and *Culex* spp mosquito populations. Depending on the species of copepod being used, other factors that can affect their use include desiccation, temperature, the presence of heavy metals, and chlorine.

Legner (1995) includes a review of the research and current knowledge concerning the use of platyhelminths (flatworms) for the biological control of mosquitoes. The advantages of their use include ease of mass production, tolerance to environmental contaminants, their ability to reproduce quickly following introduction, excellent predatory behavior in shallow-water habitats with emergent vegetation, and their ability to overwinter. Conversely, the disadvantages include the requirement that mass cultures be continuous and requires the need for highly trained technical staff, and that the persistence of flatworms in the field depends on the presence of adequate alternative food resources when immature mosquito populations are low.

2.8.1.1.2 Amphibians

Amphibians are known predators of a number of aquatic organisms including mosquitoes. Diet studies of newts, salamanders, frogs, and toads indicate that they are opportunistic generalist predators preying upon various arthropods such as cladocerans, ostracods, insects, spiders and small crayfish, as well as snails, slugs, oligochaete worms, planaria, and the occasional small vertebrate such as fish or other immature amphibians (Anderson 1968; Avery 1968; Blum et al. 1997; Brophy 1980; Bruggers 1973; Clarke 1974; Dodson and Dodson 1971; Freda 1983; Frost 1935; Fulk and Whitaker 1969; Hayes and Tennant 1985; Hamilton 1940; Korschgen and Moyle 1955; Taylor et al. 1988). Although arthropods make up a significant portion of most amphibian diets, mosquitoes appear to be insignificant as a prey item.

Specific studies and data addressing the role and/or use of amphibians as a viable and effective tool for the control of mosquitoes are limited. Howard (1901) reports on the preliminary work of a Mr. Albert Koebele, who recounted the use of salamanders that were imported from California to Hawaii. Although not a formal published study, it is one of the earliest records of attempting to use amphibians to help manage mosquito populations. Barber and King (1927) recount observations and subsequent experiments with the tadpoles of Hammond's spadefoot toad (*Scaphiopus hammondi*) in New Mexico that were found to consume crustaceans and mosquito larvae. They concluded that although the tadpoles consumed mosquito larvae, the short active season of the toad, the restriction of its habitat to temporary pools, and its lower larval consumption efficiency compared to larvivorous fishes limited its usefulness. Matheson and Hinman (1929) examined the gut contents of 59 vermilion spotted newts and noted that 47 of them contained an average of 8 or more mosquito larvae per newt. Other prey items consumed included cladocerans, ostracods, copepods, phyllopods, and an occasional aquatic insect. Feeding studies in the lab using battery jars indicated these newts were efficient predators of mosquito larvae and that when provided alternative prey preferred mosquito larvae. Matheson and Hinman were also careful to point out that in their gut content studies they did not carefully assess the full extent of the other prey items found. Spielman and Sullivan (1974) worked with tadpoles of the giant tree frog (*Hyla septentrionalis)* on Grand Bahama Island and suggested that the presence of frogs seemed to limit the abundance of the southern little house mosquito (*Culex pipiens quinquefasciatus)* in certain man-made container habitats. Their lab studies using enamel pans indicated that the tadpoles would consume mosquito larvae, especially the earlier instars, although the tadpoles did not actively pursue the larvae nor did the pans accurately reflect natural environmental conditions (e.g., other predator prey relationships that might occur as well as the presence of significant numbers of alternative prey items for the tadpoles). In large artificial containers such as 55-gallon drums and cisterns, they found tadpoles effectively controlled *Culex pipiens quinquefasciatus*. Therefore, they concluded from their lab studies, field observations of mosquito habitats with and without tadpoles, and the fact that immature mosquitoes were usually absent or in very low numbers when tadpoles were present that *Hyla septentrionalis* immatures "denied certain breeding sites to *Culex pipiens quinquefasciatus*." Freed (1980) examined prey selection, activity, and size with the Green Tree Frog *Hyla cinerea* in the lab and found that this frog consistently selected houseflies over four different mosquito species. Prey size was found to be insignificant as a determining factor for selection. Difficulty of prey capture and prey activity were significant with prey item activity being a factor in determining prey selection. Ritchie (1982) also worked with *Hyla cinerea* noting that tadpoles consumed *Culex nigripalpus* mosquito larvae and suggested that *H. cinerea* could play a significant role in the natural control of some mosquitoes in Florida. Blum et al. (1997) performed a 3-year field study of the Rhine Valley in Germany, examining the stomach contents of 2,163 anurans and found an average of 7.7 prey items per stomach with 0.16 percent consisting of mosquitoes. They concluded from their observations that the impact of anurans on mosquitoes would be negligible and, furthermore, that biological mosquito control with Bti would not negatively impact the diet of anurans. Brodman et al. (2003) worked with blue spotted and tiger salamanders in both natural wetlands and artificially created mesocosms to assess their effects on aquatic invertebrate and larval mosquito densities. They found the data from the mesocosms was consistent with what they observed in the field and that overall density of mosquito larvae in mesocosms with salamanders was 91 to 94 percent lower than those without salamanders. Therefore, the larvae of pond-breeding salamanders have the potential to control mosquitoes that use temporary wetlands. Lab studies by Willems et al. (2005) with four species of common Australian frogs found that although the tadpoles consumed some mosquito larvae, they did not consume substantial numbers and, therefore, were not effective predators of mosquito larvae. They suggested that alternate food preferences and the lack of active prey searching limited their effectiveness as biological control agents.

Brodman and Dorton (2006) examined the gut contents of 42 field-collected tiger salamanders in Indiana and found that 93 percent of all prey items observed were cladocerans and ostracods and were present in 36 percent of the stomachs dissected. Mosquitoes were found in 26 percent of the stomachs examined and comprised 1.67 percent of all prey items collected and identified from the 42 stomachs. In lab experiments, they found tiger salamander larvae could consume an average of 144 mosquito larvae per day and postulated by extension that a population of 8,000 salamander larvae could consume over 1,000,000 mosquitoes per day. Although possible, this postulation did not take into account a number of factors such as presence, abundance, availability, and seasonality of alternative prey, nor the fact that wetlands and the interactions of the organisms that reside within them are quite complex and do not readily lend themselves to such broad generalizations. Their suggestion that tiger salamander larvae could naturally reduce immature mosquito populations in wetlands seems possible, although the stomach content analysis would indicate mosquito larvae are a small portion of their overall diet. DuRant and Hopkins (2008) performed feeding experiments with red-spotted newts, mole salamanders, and the mosquitofish (*Gambusia holbrooki)*. Both the newt and the salamander readily consumed more than 300 mosquito larvae per day suggesting that these amphibians could have an impact on larval mosquito populations. The study also recognized that only a single prey item, mosquito larvae, was used and, therefore, did not account for the influence of invertebrate community composition in a natural setting on overall larval mosquito consumption rates of salamanders and newts.

A review article by Raghavendra et al. (2008) summarizes the current knowledge concerning the use of frogs for the biological control of mosquitoes. Their review notes the limited number of studies available and numerous information gaps. They also point out the need for ecological investigations that help clarify the interactions, connections and predator-prey relationships between frogs, mosquitoes, and other wetland organisms to better determine and understand the possible role of frogs as biological control agents of mosquitoes. These same concerns can also be said for salamanders, newts, and toads whose broad diets include, albeit on a limited scale, mosquitoes.

Some additional areas of concern when considering the use of amphibians as a biological control agent include, but are not limited to, (1) their inability to be used in all types of mosquito-breeding habitats (e.g., saline tidal marshes, wastewater ponds, sewage lagoons, winery waste ponds, septic tanks, storm drains); (2) what the ecosystem effects would be of introducing or mass releasing amphibians into new or nonnatural areas; (3) the ability to rear and quickly introduce large numbers of amphibians to various locations throughout the county; and (4) potential concerns by some members of the public about the sudden appearance of "large numbers" of frogs or salamanders in their yards that could also get into their homes and/or the "noise" created by large numbers of frogs in their yards at night. Although amphibians feed on a wide variety of prey items, including mosquitoes, the current knowledge and understanding indicates they have a minimal effect on mosquito populations. The District does, however, emphasize the recognition and importance of amphibians within any mosquito-breeding habitat with its staff and also makes every effort to promote the continued presence and well-being of amphibians while engaging in mosquito management activities.

2.8.1.1.3 Fish

The recommendation that certain species of fish are useful biological control agents of mosquitoes dates back to the earliest control work with mosquitoes (Felt 1904; Hildebrand 1921; Howard, 1901; Howard et al. 1912; Hardenburg 1922; Hubbs 1919; Kennedy 1916; Rockefeller Foundation 1924; Scofield 1915; Smith 1904; Stead 1907; Underwood 1903). A number of fish have been studied as potential immature mosquito predators. Walton (2007), Legner (1995), and Downs (1991) discuss the use and limitations of mosquitofish (*Gambusia* spp) as well as other fish species that have received considerable attention as potential biological control agents of immature mosquitoes. Examples of other fishes studied include, but are not limited to, the three-spine stickleback (*Gasterosteus aculeatus*), common guppy (*Poecilia reticulata*), pupfish (*Cyprinodon* spp), goldfish (*Crassius auratus*), tilapia (*Tilapia zilli*), green sunfish (*Lepomis cyanellus*), and inland silversides (*Menidia beryllina*). These and other species have been examined and in many instances found to be of limited use, not as effective as mosquitofish, or as in the cases of some fish taxa tend to outcompete the native fauna and/or are nonnatives whose use is restricted or not allowed.

Only the mosquitofish (*Gambusia affinis* and *G. holbrooki*) are commercially available for use at present, with *G. affinis* being the species typically used in the western US. Both species are very similar in behavior, biology, and habits; therefore, the term mosquitofish as used here will apply to both. It has been presumed that both species are present within California (Dill and Cordone 1997) and may even exist within the Program Area. Therefore, this review will encompass literature and data for both species.

Johnson (2008), Pyke (2005, 2008) and Swanson et al. (1996) provided a thorough review of mosquitofish biology, emphasizing information concerning physiology, growth, development, reproduction, courtship, mating, foraging behavior, diet, dispersal and movement patterns, physical and chemical tolerances, and ecosystem and interspecific interactions. Mosquitofish, in general, are quite tolerant of a wide range of environmental conditions. This feature when combined with their surface-feeding habits, ability to reproduce quickly, ease of transport, and sustainability in small volumes of water makes them ideally suited as a biological control agent of immature mosquitoes when used in carefully selected, isolated aquatic habitats (e.g., neglected residential swimming pools, ornamental ponds, water gardens, large fountains, animal troughs).

Mosquitofish, despite their name, cannot survive solely on a diet of mosquito larvae (Reddy and Pandian 1972). Laboratory and field studies have shown that mosquitofish are opportunistic omnivores that consume a wide variety of prey items, including algae, zooplankton, copepods, cladocerans, and immature stages of many insects, including, but not limited to, midges, water beetles, water boatmen, damselflies, and mayflies (Ahmed et al. 1970; Barnickol 1941; Bence 1988; Erguden 2013; Farley 1980; Garcia-Berthou 1999; Gkenas et al. 2012; Hess and Tarzwell 1942; Hildebrand 1921; Lawler et al. 1999; Mansfield and Mcardle 1998; Miura et al. 1979; Pen and Potter 1991; Reed and Hoy 1970; Rice 1941; Walters and Legner 1980; Walton and Mulla 1991; Washino and Hokama 1967). Hess and Tarzwell (1942) concluded that mosquitofish were true opportunistic feeders, so that the simple availability of prey was the key criterion in prey selection by mosquitofish. As such, the selection of food items by mosquitofish apparently shifts away from specific prey as its abundance drops.

Within their generally wide diet, mosquitofish do have some clear feeding preferences, including food at the water surface, prey size ranging from large zooplankton to very small fish or invertebrates, and prey that does not have highly effective escape behaviors (Swanson et al. 1996). While their feeding preferences, ability to rapidly reproduce and colonize a habitat, and ease of transport make these fish useful for mosquito-control purposes, their use has also generated questions and an extensive body of research concerning their potential impacts to native fauna and sensitive ecological systems

Views vary widely with respect to the beneficial and/or adverse environmental effect of mosquitofish as a biological control agent for mosquito control programs. Rupp (1996) examined 59 years of literature that contained statements pointing out the concerns of ichthyologists and some researchers about the ineffectiveness as a predator as well as the nontarget impacts to native biota of mosquitofish. Indeed, under some circumstances the generalist predatory nature of mosquitofish would make this biocontrol agent either ineffective or inappropriate to use. As with any tool used for the management of mosquitoes, proper use and placement is everything. Indiscriminate placement without regard for recognizing the sensitive nature of habitats, the diversity and density of potential prey items present at the site, the use of too few or too many fish for the size of the area, an understanding of the historical and present mosquito population dynamics, the potential for unintended relocation from the placement site, the seasonality of flooding and drying, density of vegetation, water quality, and behavior and habitat use of the managed species of mosquito are but a few of the factors that can render mosquitofish ineffective and even a potential problem for sensitive native organisms.

Rowe et al. (2008) performed an exhaustive review of the biology, behavior, and impacts of a number of invasive fishes in Australia, including the mosquitofish. Their report to the Australian government summarized the results of more than 40 studies conducted through part of 2007 concerning the impacts of *Gambusia holbrooki* to native Australian amphibians and fishes. Their review also included some of the studies conducted in the US and New Zealand concerning zooplankton and other invertebrates as well as aquatic vertebrates. This report classified the impact studies into four types: (1) correlative, using field studies on distribution and relative abundance to provide evidence that a native species may have been impacted where an introduced species now occurs; (2) impact assessments based on field studies, which use information of biology and ecology to predict the feasibility of species interactions, or tank studies, which examine the likelihood of certain interactions under controlled laboratory conditions; (3) impact assessments in the field that demonstrate the existence of impact mechanisms in the wild; and (4) species manipulations (removal of the invasive) in the field to determine if native species recover. They are careful to point out the shortcomings of each study type on their own and that it is the combination of all four that is necessary to better understand what types and range of ecosystem impacts truly are associated with the introduction of a nonnative organism. Their conclusions concerning mosquitofish are as follows: (1) irrefutable proof of their impact is lacking although a number of studies provide some evidence of impacts on native fishes and amphibians; (2) that evidence from these studies indicate that this fish can create ecological issues through a reduction in native species diversity in some areas and not in others; and (3) their ecological impact is affected by other environmental factors (e.g., temporal, spatial, weather, human-induced), which vary in intensity from location to location. The complexity of interactions between organisms within an ecosystem and, for that matter, the dynamics of any given ecosystem present many challenges when attempting to assess and understand the many biotic and abiotic inter- and intrarelationships that exist; and Rowe and coworkers plainly state this concern while reviewing the body of literature on the impacts of introduced fishes in Australia. Pyke (2008) also reviewed mosquitofish biology, ecology. and impacts and included in his discussion concerns about trophic-level effects as well as the management issues of mosquito control and wildlife management.

A study not mentioned by Rowe but of interest is that of Goodsell and Kats (1999). They performed as a part of their research a gut content analysis of 36 stream-collected *G. affinis* and found Pacific treefrog tadpoles in 65 percent of the stomachs. This result is unusual as no other gut content studies appear to have been reported with mosquitofish that co-existed with amphibians. Their laboratory and field experiments also showed that mosquitofish preyed upon treefrog tadpoles even when high densities of mosquito larvae were given as alternate prey.

Mosquitofish impact studies on frogs and fish since Rowe et al. (2008), as well as all research on other organisms also neatly fits into their categorization system of correlative, two types of impact assessment and, invasive species manipulations or removal. Most of the research continues to be either correlative or impact assessments that demonstrate the likelihood of certain interactions under controlled laboratory conditions. The remaining discussion will review research on frogs and fish since Rowe and coworkers report and then review the research both before and after Rowe for other amphibians and wetland organisms.

The interactions between mosquitofish and frogs have begun to receive more attention with the increased awareness that most research had been focused on mosquitofish interactions with native fishes. Gregoire and Gunzberger (2008) used tanks in the lab to assess the effects of three species of predatory fish, including *Gambusia holbrooki*, on the survival and behavior of the southern leopard frog and the gopher frog. Their observations noted that mosquitofish did injure the frog tadpoles, which increased tadpole hiding behavior. They suggested that the introduction of predatory fish could negatively affect frog populations especially in normally fish-free wetlands. Note that this study did not provide alternative prey and, therefore, focused strictly on the interactions between the fish and frog tadpoles contained within 10-gallon aquaria. Karraker et al. (2010) worked with four species of frogs and one toad from the lowland wetlands of southern China and noted that the four species of frog tadpoles were susceptible to predation. They also suggested that the other frog species present within the Chinese lowland wetlands may be subject to predation and, therefore, further investigation and potential conservation measures should be taken. It is of interest to note that predation testing in this study occurred in small containers, and no form of refugia was provided for the tadpoles. Stanback (2010 observed the interaction of hatchling tadpoles of the upland chorus frog (*Pseudacris feriarum)* and mosquitofish in 100-gallon cattle tanks. Those tanks with fish had no tadpoles remaining while the tadpole-only tanks had approximately 10 percent of the introduced tadpoles still present. He suggested that mosquitofish were highly effective predators of hatchling tadpoles, even of frogs that have co-evolved with this fish. This particular study does have some issues though. First, a high density of fish was placed into the tanks. Second, no assessment was done concerning the presence or abundance of alternative prey. Third, as Stanback pointed out, the potential for tadpole cannibalism was not taken into account.

The study by Shulse et al. (2013) is different from most prior research in that they examined mosquitofish and the development of community structure over a 4-year period in constructed experimental wetlands. They found that the introduction of mosquitofish reduced the abundance of two species of grey treefrogs (*Hyla versicolor* and *H. chrysoscelis*), the boreal chorus frog (*Pseudacris maculata*), and aquatic invertebrates. They also noted that mosquitofish had no significant effect on the green frog (*Lithobates clamitans).* When mosquitofish were removed, invertebrate abundance increased; and they suggested that mosquitofish removal may have also been a contributing factor in chorus frog recolonization of the experimental wetlands with low invertebrate predator abundance. From their observations, they suggested that mosquitofish were detrimental to wetland communities and recommended against their future use.

Reynolds (2009) performed a gut content, field correlative, and lab-controlled species interactive study and essentially found minimal impact when assessing the effect of *Gambusia holbrooki* on six species of amphibians in Southwestern Australia. His study noted that the gut contents of 48 fish collected from five wetland sites where frogs occurred showed no evidence of frog eggs or tadpoles. Furthermore, in laboratory feeding trials, mosquitofish that had not fed for 4 days did not consume the eggs of any of the frog species worked with in the lab but did consume alternate invertebrate prey at the end of the egg palatability trials. Unfortunately, trials with tadpole hatchlings for all but one species and the older tadpoles of one frog species did result in a mosquitofish-feeding response. Reynolds also surveyed 25 wetland sites and found mosquitofish at 20 of the sites, frogs at all 25 sites, and that the combination of all frog species co-existed with fish at more than 8 of the sites. Most importantly, he noted that frog species richness did not differ between sites with and without mosquitofish. He concluded that "in contrast to the situation in eastern Australia, populations of anuran species in southwestern Australia do not appear to be strongly affected by this small invasive fish." It was further suggested that other factors such as frog egg deposition site, breeding time of the frogs, availability and abundance of alternative prey, condition of the wetland, and temporal variation in fish abundance and size may influence the impact of mosquitofish on frogs. Bottom line, the interaction of mosquitofish and amphibians is a complex issue and requires careful consideration and analysis in light of the many studies completed to this point in time.

Two additional studies of note are those of O'Meara and Darcovich (2008) and Alvarez et al. (2004), both of which report changes in frog populations following the removal of nonnative fish. Alvarez et al. (2004) surveyed 90 managed stock ponds within Kellogg Creek Watershed (Contra Costa County, California) and noted 7 ponds with exotic fish, 4 of which also had mosquitofish, and that these ponds had little use as well as almost no reproduction by red-legged frogs. Two of the ponds in particular only had large populations of mosquitofish. When the fish were removed, frog use and reproduction improved. O'Meara and Darcovich (2008) report the increase of green and golden bell frogs following the rotational drainage and subsequent removal of mosquitofish from ponds in a wetland park that had 22 habitat ponds constructed for the frog. Over a period of 3 years, a different set of ponds, or about one-third of the 22 ponds, were drained and allowed to remain dry for a period of 4 weeks. Ponds were then refilled and monitored. Although fish and frogs did coexist, frog numbers, especially of tadpoles and juveniles, improved with the reduced presence of mosquitofish. Additionally, sightings of adult frogs were significantly improved and had reached their highest levels on record since construction of the wetland in 2000, in 2004–2005, and again in 2005–2006. Note that fish did reinvade many of the ponds within a few months of draining, and data concerning tadpole numbers post-fish recolonization are not clearly presented. Also, data are lacking concerning the presence and abundance of other potential predators of these frogs with the exception of a passing reference to a predatory eel.

The more recent impact studies concerning native fish are varied both in scope and design although one attempted to assess the removal of mosquitofish in the field to determine if native species would recover. Laha and Mattingly (2007) observed the interaction between *Gambusia affinis* and the barrens topminnow (*Fundulus julisia)* in small glass tanks in the lab. They noted that for short-term exposures, juvenile topminnows were quite vulnerable to aggression and predation, which the authors attributed to mosquitofish. A 60-day interaction study with the adults of both species yielded no negative effects with the exception of fin injury to top minnows that were syntopic with mosquitofish. From their observations they suggested that the impacts of mosquitofish on barrens topminnows was primarily through predation and injury to the early life stages. Laha and Mattingly also pointed out the limitations of their experimental design, specifically that not all dimensions of the natural environment could be adequately represented. Keller and Brown (2008) examined the behavioral interactions that occurred between allopatric and sympatric populations of wild-caught *Gambusia holbrooki* and the native Australian ornate rainbowfish

(*Rhadinocentrus ornatus)* in laboratory-maintained aquaria and from their observations suggested that mosquitofish presence and aggression was responsible for the behavioral and microhabitat shifts that occurred with the rainbowfish. They noted that rainbowfish individuals from allopatric populations were more susceptible to fin nipping and being chased than their sympatric counterparts, that sympatric rainbowfish had shifted their microhabitat preferences (thus allowing them to coexist with mosquitofish), and also exhibited a greater level of aggression during all stages of mosquitofish exposure. MacDonald et al. (2012) performed a quantitative survey of 93 wetlands in southeastern Australia in an effort to develop a model of the influence of *Gambusia holbrooki* on native fish species diversity, abundance, and physical condition. From their findings, they asserted that *Gambusia holbrooki* exerted a strong effect on the likelihood of wetlands being occupied by most other species of fishes and that their level and direction of influence on the presence, abundance, and/or physical condition of different fish taxa seemed dependent on both biotic and abiotic factors. They also observed that three species of native fishes, Australian smelt, flat-headed gudgeon, and carp-gudgeon, co-existed with mosquitofish in wetlands without aquatic vegetation and both species of gudgeon juveniles showed no strong evidence of fin damage. Therefore, they proposed that some generalist life-history strategies may insulate certain native fish species allowing them to successfully coexist with mosquitofish. Tonkin et al. (2014) performed a field based study, using 13 wetland sites, to assess the effects of mosquitofish removal on wetland fishes. Like MacDonald (2012), a predictive modeling approach was used and overall they found that three species of fish, carp gudgeon, Australian smelt, and common carp, did not respond to removal of mosquitofish. They suggested that the limited duration of the study and number of sites and possibly even the species used may have played a role in the inability to detect mosquitofish's impact on the rate of population change for any of the three species of observed fish. However, the authors did conclude that their data supported the earlier findings of Macdonald et al. (2012) that suggested the direction and level of impact of mosquitofish on wetland fish species was quite fluid and dependent on biotic and abiotic influences. They also suggested that those organisms with more flexible life-history strategies are more able to coexist with mosquitofish than those that overlap more in time, diet, and habitat.

Other studies with mosquitofish suggest they may impact some species of native salamanders and newts, aquatic invertebrate populations, and planktonic communities. Gamradt and Kats (1996) surveyed 10 streams in the Santa Monica Mountains of Southern California in 1994 and 1995 and compared those data to other surveys conducted between 1981 and 1986. All streams contained newts in the 1980s surveys. The 1994–1995 surveys found three streams no longer had newts but did have mosquitofish and/or crayfish. Furthermore, the remaining seven streams that had newts did not have the introduced predators mosquitofish or crayfish. Their subsequent lab studies using small tubs found that mosquitofish would consume larval newts but not the eggs. This correlation study though has a significant limitation. No way exists of really knowing that mosquitofish were responsible for newt disappearance in the three streams. Many other possible factors, biotic and abiotic, could be responsible for newt absence (e.g., human influence on water flows, unknown released contaminants, disease).

Leyse and Lawler (1998, 2000) investigated the relationship between mosquitofish and the California tiger salamander (*Ambystoma californiense),* in six 3.05 x 6.1 x 0.6-meter outdoor experimental ponds. The results of their 1998 experiment showed that mosquitofish presence did not affect either larval growth, weight, size, or the number reaching metamorphosis. Their trials with aquaria in the lab indicated young salamander larvae often successfully swam away from mosquitofish when attacked, although a few were consumed. Their second set of pond experiments yielded different results and they reported delayed metamorphosis, tail injuries, decreased weights, and significant reductions in salamander survival. Note that the significant difference between the two experiments was the initial number of fish stocked in the ponds between the two trials. The first trial used 12 fish per pond (placed in February) while the second stocked 300 fish per pond. Mosquitofish populations drop to a very low level during the winter months as most adult females and nearly all adult males die during this time period (Swanson et al. 1996). Therefore, when activity begins in the spring very few adults remain and they serve as the population that will begin reproduction, which occurs in late spring. Large numbers of fish are typically not observed until well into summer. The higher initial February stocking rate was well beyond what would normally be seen for winter survival and, therefore, altered the temporal and spatial separation that would otherwise have occurred between salamanders and mosquitofish populations. Segev et al. (2009) while observing fire salamander (*Salmandra infraimmaculata*) and *Gambusia affinis* populations in three natural pools noted that salamander larvae exhibited damage consistent with mosquitofish biting activity when mosquitofish populations were high. They also found the tail:body ratios of the salamander larvae were significantly higher (longest tails) when mosquitofish were absent. When comparing observations for the years 1999 and 2003, which were pre- and post-introduction of mosquitofish for two of the pools, they also found larval salamander densities had significantly decreased. Their mesocosm experiments using 180-liter containers in the lab demonstrated results similar to what they observed in the natural pools, that is mosquitofish-impacted larval salamander survival and size, and fish attacks resulted in damaged appendages.

Invertebrate and planktonic interactions with mosquitofish vary although again it has been suggested from the data and observations that potential impacts may occur. Leyse et al. (2004) tested the effects of mosquitofish on the fairy shrimp (*Linderiella occidentalis)* in experimental ponds and found fairy shrimp survival and invertebrate biomass was significantly reduced in ponds with mosquitofish. Their feeding trial studies with lab aquaria also demonstrated that mosquitofish generally preferred fairy shrimp to alternative prey. From their data, they suggested that mosquitofish introductions into naturally fishless wetlands could impact species diversity. Mosquitofish interactions with insects indicate that reductions can occur (Bence 1982; Farley and Younce 1977) and may be influenced by the seasonal nature of mosquitofish population density, fish size (which affects prey selection), instar of the insect, and stocking rate (Bence 1982; Miura et al. 1984; Walton and Mulla 1991). Planktonic studies have also shown declines in abundance as well as shifts in population structure (Bence 1988; Bence and Murdoch 1982; Hurlbert and Mulla 1981; Margaritoria et al. 2001; Singh 2013). Of particular interest is the study by Singh (2013) who also found higher pH, dissolved oxygen, and turbidity in ponds following introduction of mosquitofish. Singh concluded that mosquitofish demonstrated a top-down effect on zooplanktonic and phytoplantonic community structure and abundance and, therefore, could be one of the factors affecting the productivity in Lake Nainital, which had received mosquitofish as part of a mosquito control program in the 1990s.

From the review of the above research, it is clear that mosquitofish do have the potential to impact the environments within which they are placed. Yet, it is also important to remember to be careful when working with and evaluating the data from the myriad of studies that have been conducted with mosquitofish. While these studies suggest that mosquitofish can reduce populations of amphibians, fish, and invertebrates, some significant factors need consideration when working with the data:

> *First*, the results of many of these studies are laboratory based and use artificial environments that are limited in their ability to emulate natural fully functioning wetland habitats and/or they offer the fish limited prey selection.

> *Second*, some studies created outdoor simulated wetland mesocosms; yet even these sites were limited as they did not in many cases have the diversity of microhabitats, vegetation, and full range of complex biotic interactions that one might find in well-established natural wetland systems.

> *Third*, many studies lacked populations of potential predators of mosquitofish that can be found in natural habitats, thus allowing the populations of mosquitofish to exceed levels that would otherwise be found.

> *Fourth*, some studies use stocking rates well above rates used by mosquito control agencies or had stocked ponds with numbers of fish that were much higher than what would occur in the wild for that time of year.

Walton (2007) cogently points out that "*predation on mosquitofish, environmental complexity and environmental factors may ameliorate the strong effects observed in simple laboratory and mesocosm systems*."

Although little doubt exists that mosquitofish are a useful biological control agent of immature mosquitoes, their use and application does have limitations both in terms of effectiveness and in reducing the risk of potential unwanted impacts. The District supports and encourages the presence of the other biological control predators of mosquitoes when practical. Yet, the only readily available biological control agent for use, other than the bacterium *Bs*, is the mosquitofish. The rearing and stocking of mosquitofish in mosquito-breeding habitats is also the most commonly used biological control agent for mosquitoes in the world. Nonetheless, the District's use of mosquitofish is limited and carefully monitored and includes rechecking planted sites to verify presence and abundance. Mosquitofish are typically used as a long-term control measure and, therefore, are not planted in habitats prone to drying. Fish are placed in closed man-made water features such as ornamental ponds, water gardens, horse troughs, rainwater barrels, and large fountains, and care is taken to verify that this biological control agent cannot gain access to unintended habitats, especially creeks and sensitive wetlands. Citizens are also advised of California State regulations prohibiting the introduction of nonnative species (e.g., mosquitofish) into waters of the state and the US. Operationally, the use of mosquitofish is also limited by factors such as highly polluted water (e.g., dairy lagoons, winery waste ponds, septic ponds), presence or proximity of sensitive species and habitats, and whether or not the mosquito-breeding site is a seasonal water source or a permanent impoundment.

Therefore, the District, as mentioned above, limits the use of mosquitofish to those sites with reduced potential for impacts to native species and sensitive habitats to occur. Mosquitofish are stocked only in compliance with federal and California endangered species regulations, so as to avoid the potential to harass and impact threatened and endangered fish, amphibians, insects, and other wildlife. District staff are highly trained, are certified by the California Department of Health Services, and are required to complete frequent continuing education sessions sponsored by the state, the District, or the Mosquito and Vector Control Association of California. Lastly, District staff routinely coordinates and consults with other responsible agencies, including the California Department of Health Services, California Department of Fish and Wildlife, US Fish and Wildlife Service, San Francisco Bay Conservation and Development Commission, California State Lands Commission, San Francisco Regional Water Quality Control Board, US Army Corps of Engineers, Napa County Resource Conservation District, and Napa County Flood Control District to ensure that biological control activities with mosquitofish do not result in substantial adverse effects to biological resources.

2.8.1.1.4 Bats

The concept of bats as effective predators of mosquitoes has been espoused for the better part of 100 years (Campbell 1925; Felt 1904: Grinnell 1918; Howard 1901; Howard et al. 1912: Underwood 1903). The biggest surge in this theory came about with the assertions of Dr. Charles Campbell, who had been publicly sharing his work with bats and the construction of bat houses near wetlands for many years prior to the release of his 1925 publication entitled *Bats, Mosquitoes and Dollars*. His work received wide acclaim and became highly popularized with the general public. Unfortunately, the concerns of scientists with contradictory data from consulted experts, or who were themselves knowledgeable about such matters, went unnoticed (Goldman 1926; Howard 1920; Nelson 1926; Storer 1926). The link between bats, the construction of bat houses near wetlands, and numbers of mosquitoes affecting people had not been made as no definitive data were presented from the examination of fecal pellets or gut contents. Ross (1967) provides a summary of the work examining Dr. Campbell's claims.

Fifteen species of insectivorous bats within the families Molossidae and Vespertilionidae are known to occur within the San Francisco Bay region (see: www.sfbaywildlife.info/species/mammals). The Molossid bats are the Brazilian (Mexican) free-tailed bat (*Tadarida brasiliensis*) and the western mastiff bat (*Eumops perotis*). The Vespertilionid bats are the pallid bat (*Antrozous pallidus*), big brown bat (*Eptisicus fuscus*), silver-haired bat (*Lasionycteris noctivagans*), western red bat (*Lasiurus blossevillii*), hoary bat (*Lasiurus cinereus*), California myotis (*Myotis californicus*), long-eared myotis (*Myotis evotis*), little brown myotis (*Myotis lucifugus*), fringed myotis (*Myotis thysanodes*), long-eared myotis (*Myotis volans*), Yuma myotis (*Myotis yumanensis*), Townsend's big-eared bat (*Plecotus townsendii*), and western pipistrelle

(*Pipistrellus hesperus*). Published research shows that some of these bats do indeed feed on mosquitoes, although the assertion that mosquitoes are a significant or primary part of their diet, which therefore makes them effective predators and by extension good biological control agents of mosquitoes, does not always hold true.

Published dietary studies examining gut contents or the guano of bats within the San Francisco Bay area appears limited. A number of observations and studies though from other parts of California and throughout the US suggest flies, especially mosquitoes, constitute a small portion of the diet for most of these bats (Black 1974; Borell 1942; Brigham and Saunders 1990; Buchler 1976; Easterla and Whitaker 1972; Feldhamer et al. 2009; Freeman 1979; Hamilton 1933; Hatt 1923; Kunz et al. 1995; Orr 1954; Perlik et al. 2012; Ross 1961, 1967; Whitaker and Barnard 2005; Whitaker 1972; Whitaker and Lawhead 1992; Whitaker et al. 1981a,b; and Whitaker et al. 1996). Midges, belonging to the family Chironomidae have been found in both gut content and fecal pellet analysis to be a significant part of the diet of the little brown myotis and the California myotis in western Oregon (Whitaker 2004; Whitaker et al. 1977). Additional studies by Anthony and Kunz (1977) and Belwood and Fenton (1976) also show aquatic insects, especially midges, are important to the little brown myotis' diet.

Although gut content and fecal analysis help to confirm and clarify the diets of these bats, it is important to be careful when working with and interpreting these data. The different regions where these studies and observations occurred, the insect fauna available as prey items at these locations, and time of year the studies were conducted are but a few of the factors that can account for the variability in diet and behavior of these species of bats. Existing data would indicate that overall, bats may consume some mosquitoes but without further research it is hard to claim bats are an effective biological control agent of mosquitoes. However, the District does make every effort to provide information to interested individuals and organizations about bat conservation.

2.8.1.1.5 Birds

Insectivorous birds are another of the many potential predators of mosquitoes within Bay area wetlands. The San Francisco Bay region supports a number of species of insect-eating birds; however, it does not appear that any feed exclusively on mosquitoes. Some of these species (e.g., purple martins, swallows) have received considerable promotion as effective predators of mosquitoes that could, with the erection of nesting boxes and achievement of sufficient population levels, significantly reduce the need for other forms of mosquito control, especially chemical sprays (http://www.birdnote.org/show/barn-swallow-natural-pest-control; Blickle 2011; http://www.makeyourownbirdfood.com/mosquito.html; http://www.ruralsurvival.com/mosquito_control.html; Wade 1966). Unfortunately, feeding observations and gut content analysis studies of these opportunistic insectivores do not support the anecdotal claims for purple martins and swallows as effective biological control agents of mosquitoes (Beal 1918; Farley 1901; Grant 1945; Jackson and Weber 1975; Johnston 1967; Mengelkoch et al. 2004; Orlowski and Karg 2011; Riggs 1947; Walsh 1978).

Kale (1968) reviews purple martins and their effectiveness as a biological control agent of mosquitoes. He discusses (as mentioned above) that gut content analysis studies indicated mosquitoes were an insignificant part of their diet. He further pointed out that these birds typically fly from 100 to 200 feet above the ground, although they can and do fly anywhere from a few inches to an altitude of almost 500 feet. Since mosquitoes tend to remain closer to the ground (below tree canopy) and are usually active during hours that purple martins typically are not, mosquitoes are significantly limited as a viable prey item for these birds. Kale goes on to review literature that makes unsubstantiated claims concerning purple martins and mosquitoes. Of special interest are Wade's (1966) claims, wherein purple martins could conservatively consume 2,000 mosquitoes per day and when mosquitoes are plentiful up to 10,000–12,000 per day. Kale points out that not only is this claim misleading, it is without any basis in fact. Grossman (1990) reiterates Kale's point and also references another article by Hill (1989) which discusses a number of purple martin myths including their effectiveness as a mosquito predator. Wiggins

(2005) provides a conservation assessment that includes a thorough and detailed review of the exiting literature concerning purple martins. The reader is referred to this document for additional information on biology, ecology, information gaps, and recommended management practices. Though more specific for the Great Plains and the Rocky Mountain region, still a great deal of useful information is presented on this bird species.

Studies on foraging ecology, prey size, and selection of swallows indicate a preference for smaller prey items, especially small flies (McCarty and Winkler 1991, 1999; Quinney and Ankney 1985; Turner 1982). This preference would suggest that mosquitoes, when available, would be opportunistically used as a food source. Unfortunately, some species of mosquitoes are active during periods when swallows are inactive. In other instances, mosquito species' seasonal abundance does not fully coincide with that of swallows. Lastly, some mosquito species breed within habitats and areas that swallows are unable to readily access and consume them.

The consideration and use of insectivorous birds also has legal constraints. At the federal level, Title 16, Chapter 7 protects migratory game and insectivorous birds. Migratory Bird Treaty Act Sections 701 and 702 clearly state that it is unlawful to take, possess, import, export, transport, purchase, barter, or offer for purchase, barter or sale, any migratory bird, their eggs, parts, and nests except with a valid permit. Exceptions are limited but local governmental mosquito and vector control agencies are not included. Title 50, Section 10.13 contains the list of migratory birds the act protects. Also note that some of the species on this list also appear on the list of endangered and threatened species under Title 50, Section 17.11, Endangered Species Act. At the California state level similar restrictions apply, although permits can be obtained for specific scientific studies and research pursuant to any endangered and threatened species limitations or sensitive habitat concerns. Therefore, if birds were to be an effective biological control agent of mosquitoes, and they were also available and could be used, then the additional limitations to be addressed would include (1) managing the bird population to minimize unintended impacts to other native fauna, (2) making sure sufficient habitat and resources exist for optimal survival of the released birds, (3) making certain the birds are disease free at the time of release and do not unintentionally introduce pathogens into native bird populations, and (4) ensuring the release does not significantly increase the potential population of reservoir organisms for mosquito-borne pathogens.

Kale (1968) points out an additional significant factor concerning the potential of insectivorous birds serving as an effective biological control agent, specifically that even when insects are abundant birds remove a very small proportion. He references the works of Lack (1967) and Andrewartha and Birch (1961) noting that the timing of the abundance of insects tends not to coincide with the timing of maximum bird population presence. Therefore, the numbers of birds needed are insufficient to effectively control high pest insect densities. From a historical perspective, the anecdotal accounts of mosquito abundance in the diaries of early settlers and explorers (Bolton 1927; Gray 1951) would seem to bear this out as the wetlands at the time were pristine and contained abundant populations of birds, bats, and mosquitoes.

The potential for the spread of mosquito-borne disease, especially West Nile virus, is a concern that needs more research, especially as it relates to birds and mosquito control. Both the Centers for Disease Control and Prevention (CDC) and the US Geological Survey maintain lists of those birds that have tested positive for the presence of West Nile virus. McLean (2006) discusses the impact of West Nile virus on the North American bird fauna noting that more than 200 different species of birds have tested positive. Although not all birds readily succumb to infection or make for good reservoir hosts, much is still being learned about West Nile virus and its impacts to birds as well as the amplification of this virus in different bird populations. Wheeler et al. (2009) reviews California bird surveillance data from 2004 to 2007 and notes that bird susceptibility varies widely. Oesterle et al. (2009) experimentally infected cliff swallows with West Nile virus and suggested that cliff swallows are a competent reservoir host and could, therefore, play a role in the early season amplification and maintenance of West Nile virus. Since this virus is still relatively new to the North American continent, exercise caution, as much is still unknown about the relationships of this virus and North American birds.

Therefore, intentionally breeding and mass releasing birds to help manage mosquito populations is not viable at this point in time and could have adverse effects on the ecosystem, including the epidemiology of West Nile virus. The lack of hard data to support the claims of effective mosquito predators, the potential for mosquito-borne disease issues, and regulatory constraints are but a few of the factors limiting the use of insectivorous birds as a biological control tool. However, the District does make every effort to support and encourage the health and well-being of potential mosquito predators, by providing information about bird biology, ecology, and conservation.

2.8.1.2 Yellow Jacket Predators

Effective predators that specifically target yellow jackets are apparently unknown. Opportunistic predation of yellow jackets does occur and is usually the result of animals such as raccoons and skunks that have happened upon and then dig up a nest. Popular media (e.g., on the internet) have suggested that placing attractants such as honey around the entrance of the nest to attract raccoons or skunks is an effective means of eliminating a yellow jacket nest (Beyond pesticides.org, downloaded July 2014). Unfortunately, raccoons can cause unwanted effects (e.g., getting into refuse cans, attacking pets, digging up vegetable gardens) not only to the property with the nest but potentially to neighboring properties as well. Skunks, of course, may leave an unwanted odor as a result of becoming alarmed by pets or other unexpected disturbance while they are foraging. Therefore, this method of management and control is not encouraged or recommended.

2.8.2 Examples of Tool Use

Examples of mosquito predators include representatives from a wide variety of taxa: coelenterates, *Hydra* spp; platyhelminths, *Dugesia dorotocephala, Mesostoma lingua, and Planaria* spp; cyclopoid copepods; insects, *Anisoptera, Zygoptera, Belostomidae, Geridae, Notonectidae, Veliidae, Dytiscidae, Hydrophilidae, and mosquitoes (Toxorhynchites* spp*)*; arachnids, *Pardosa* spp; fish *Gambusia affinis*, *Gasterosteus aculeatus*; bats, Molossidae and Vespertilionidae; and birds, anseriformes, apodiformes, charadriiformes, and passeriformes. Examples of yellow jacket predators include, but are not limited to, raccoons, skunks, and potentially a few species of wasps and spiders. Examples of rodent predators include, but are not limited to, cats, hawks, falcons, owls, skunks, raccoons, and some species of snakes. Examples of tick predators include, but are not limited to, a few species of: spiders (*Dysdera murphyi*, *Teutana triangulosa*, wolf spiders), mites, assassin bugs, ants, beetles (carabidae, cantharidae and silphidae), toads (*Bufo paracnemis*), lizards, and birds (charadriiformes, ciconiformes, coraciformes, cuculiformes, galliformes, tinammiformes, and passeriformes). It is important to note that none of the predators listed above, except the mosquitofish (*Gambusia affinis*), are commercially available for large-scale biological control programs.

2.8.3 Applicability to District IMMP

The District does use mosquitofish to manage certain domestic mosquito populations. Fish are typically placed in closed man-made water features such as ornamental ponds, water gardens, horse troughs, rainwater barrels, large fountains, and abandoned swimming pools. Their use is limited, carefully monitored, and restricted to those sites with reduced potential for impacts to native species and sensitive habitats. Citizens are also advised of the regulations prohibiting the introduction of nonnative species (e.g., mosquitofish) into waters of the state and the US.

Other mosquito predators such as coelenterates, platyhelminths, copepods, aquatic insects, spiders, bats, and birds are not commercially available for use at this time. Therefore, the District makes every effort to encourage the presence of any natural predators that are already present within mosquito-breeding habitats.

No commercially available predators of yellow jackets exist at this time. Therefore, this vector management tool is not available for use as a part of the District's IMMP.

2.9 Biological Control Plants

2.9.1 Description

Insectivorous plants have been mentioned as possible biological control agents of mosquitoes for more than 100 years (Howard et al. 1912; Matheson 1932). One genus of particular interest has been the bladderworts (*Utricularia* spp), an aquatic plant that has been studied sporadically for more than 80 years. Other plants include, but are not limited to, species such as pitcher plants (*Sarracenia* spp), sundews (*Drosera* spp) and Venus fly trap (*Dionaea muscipula*). The District is not aware of any native carnivorous plants within the Program Area, although some citizens cultivate carnivorous plants, especially *Sarracenia* spp.

Matheson's (1931) review of aquatic plants and mosquito control summarizes both previous and current work with bladderworts and concludes that these plants deserve further consideration as possible biological control agents of mosquitoes. He also points out that food items were varied, consisting of small crustaceans, protozoa, and even some small insects, including immature mosquitoes. Angerilli and Bierne (1974) observed the influence of certain freshwater plants, including *Utricularia minor*, and noted significant but inconsistent levels of mosquito larval predation. Unfortunately, this study placed these plants in tanks that used tap water, which also did not have significant populations of alternative prey items. Baumgartner (1987) studied bladderworts and their effectiveness as a predator of late fourth instar *Culex pipiens* larvae in the lab and determined that these carnivorous plants do capture mosquito larvae but are inefficient as a biological control agent of mosquitoes. Gordon and Pacheco (2007) examined prey composition of two bladderwort species in Venezuela and found prey items consisted of rotifers, cladocerans, copepods, rhizopods, annelids, insects, and various phytoplankton. Note that algae made up more than 60 percent of prey items while animal forms were the remainder (insects comprising an insignificant amount of the animal taxa consumed). Ogwal-Okeng et al. (2011) worked with *Utricularia reflexa* and another species of carnivorous plant to test their larvicidal activity on Anopheles mosquitoes and concluded that *Utricularia reflexa* significantly reduced mosquito populations and was, therefore, a potential biological control agent for control of mosquitoes and malaria in Uganda. Again it should be pointed out that the small tanks used in this study did not accurately reflect natural environmental conditions (e.g., the presence of significant numbers of alternative prey items).

The terrestrial *Sarracenia* spp seems to be even less effective as predators of mosquitoes. Cresswell (1991) examined the insect prey of 214 pitchers of the pitcher plant (*Sarracenia pupurea*) for a period of 55 days. A total of 504 prey items were extracted (503 belonging to 49 families of insects), and only 4 prey items were mosquitoes. Wray and Brimley (1943) collected the prey and inquilines of *Sarracenia flava*, *S, purpurea,* and *S. rubra* from five localities in North Carolina, two of the sites being well populated by pitcher plants and a quarter acre in size or larger. With the exception of the larval forms of *Wyeomyia smithii*, a species of mosquito whose larvae develop in the water of the pitcher plant, four adult mosquitoes were found as prey items. The popular idea that large numbers of pitcher plants prevent human interactions with mosquitoes seems dubious and has yet to be proven beyond what is found anecdotally.

Sundews also have a wide range of invertebrate prey, although prey composition and capturing efficiency varies depending on the sundew species involved (Baltensperger 2004; Thum 1986). Other than anecdotal information, a paucity of scientific research appears to examine the efficiency of sundews as a biological control agent of mosquitoes.

2.9.2 Examples of Tool Use

The District does not employ the use of carnivorous plants.

2.9.3 Application to District IMMP

Carnivorous plants, whether terrestrial or aquatic, use a wide range of invertebrate prey and are not specific predators of mosquitoes. What little data exist indicate that carnivorous plants, especially terrestrial species, are inefficient for the control of mosquitoes. Therefore, the District does not use carnivorous plants as a part of its IMMP.

2.10 Synthetic Insecticides

2.10.1 Description

The District sometimes uses synthetic insecticides such as the pyrethroids etofenprox and resmethrin, as well as the organophosphate temephos to help manage mosquito populations. These active ingredients were evaluated in the Ecological & Human Health Assessment Report prepared for the PEIR as Appendix B. This report evaluated the potential hazards and estimated risks of these and other active ingredients (in Section 4.1). Documented toxicity and environmental fate of these insecticides were reviewed and evaluated. Those that demonstrated reasonable efficacy with minimal undesirable effects (i.e., unwanted estimated risk) were selected for inclusion in the District's IMMP.

The use of synthetic insecticides is the least preferred method due to the potential impacts to nontarget organisms. All of these materials are highly effective but nonselective. Therefore, this tool is used when all other methods are no longer practical or have not been successful in effectively reducing mosquito populations to a healthful level for people, pets and livestock.

2.10.2 Examples of Tool Use

Etofenprox, Temephos, Resmethrin

2.10.3 Applicability to District IMMP

The District currently uses etofenprox as a mosquito adulticide. The method of application is restricted to ultra low volume (ULV) fogging using truck-mounted, all-terrain vehicle (ATV)-mounted, and handheld fogging machines. The organophosphate temephos (used in solid granular form only) has been used in the past and although there are currently no plans to use it in the future, the District reserves the right to use it in order to combat resistance. Use would be limited to areas where the potential for introduction to unintended habitats and exposure to nontarget organisms can be effectively managed. Resmethrin is a tool that could be used in the event that etofenprox was not available for a planned adulticiding application. Therefore, resmethrin is a tool for potential future use by the District.

2.11 Natural Insecticides

2.11.1 Description

Natural insecticides are those materials made directly from plants (botanical insecticides) or other organisms such as bacteria. Some of these materials, such as Bti, are highly host specific, while others such as pyrethrin are not.

2.11.1.1 Botanical Insecticides

Botanical insecticides (e.g., pyrethrin) are derived from natural plants in contrast to the synthetic versions described in Section 2.10. Pyrethrin (pyrethrum) is one of the most commonly produced and used natural insecticides and is sometimes used by the District as a part of its IMMP. Pyrethrin is a natural insecticide extracted from certain varieties of the flower *Chrysanthemum cinerariaefolium* and consists of six active ingredients collectively known as pyrethrins (EPA 2006; Gunasekara 2005; Worthing and Hance 1991). This insecticide provides effective control of adult mosquitoes and other insect pests at very low dosage and has little residual activity (persistence) due to its sensitivity to sunlight. The chrysanthemum flowers used to produce pyrethrins are grown commercially in parts of Africa, Asia, and Australia.

Pyrethrins and pyrethroids exhibit rapid knockdown and kill of adult mosquitoes, characteristics that are considered a major benefit of their use. The mode of action of these compounds relates to their ability to affect sodium channel function in the insects' neural membranes. Their toxicity in insects is markedly increased by the addition of synergists (primarily piperonyl butoxide) which inhibit detoxification of the pyrethrins in insects. No evidence suggests that these synergists increase toxicity in mammals.

Pyrethrins are not cholinesterase inhibitors, are noncorrosive, and will not damage painted surfaces. They are less irritating than other mosquito adulticides and have a less offensive odor. In comparison to other adulticides, pyrethrins may be effectively applied at much lower rates of active ingredient per acre.

The District recognizes that pyrethrins can impact other organisms, especially insects, and therefore takes great care when using this insecticide to minimize effects to nontarget organisms. At mosquito control label application rates, aerial spraying with pyrethrins showed no impact on large-bodied arthropods but did have some impact on small-bodied organisms (Boyce et al. 2007). Jensen et al. (1999) found that ULV applications over three seasonal wetlands on Sutter National Wildlife Refuge resulted in no detectable reduction in the abundance or biomass of aquatic macroinvertebrates. However, they did find a temporary decrease in flying insect abundance in both treated and control wetlands that recovered within 48 hours. Davis et al. (2007) performed an ecological risk assessment of pyrethrins, permethrin, resmethrin, phenothrin, and two organophosphates to warm- and cold-water vertebrates and aquatic invertebrates that may be in a watershed where mosquito spray applications occur, as well as those mammals and birds that would be in the spray zone. They found that the risk quotient for pyrethrin was low, suggesting that the risk to nontarget receptors was most likely small.

The District sometimes uses pyrethrin to manage adult mosquitoes and potentially yellow jackets. The use of pyrethrin is also a least preferred method for controlling mosquitoes. Operationally, the District is very careful when using pyrethrins, as they are not selective for mosquitoes. Therefore, use near beehives is restricted. Additionally, wind restrictions also apply to minimize unwanted drift when making ULV fogging applications.

2.11.1.2 Bacterial Insecticides

Insecticides derived from bacteria (e.g., Bti and *Saccharopolyspora spinosa*) typically consist of a chemical byproduct and/or protein spore produced directly from the organism. These materials tend to be highly effective for managing the immature stages of mosquitoes, are nonpersistent, and, depending on the formulation, can be relatively target specific. The application of these materials is labor intensive and requires proper application and full access to the immature mosquito-breeding sites to be effective.

The bacterium Bti produces spores containing protein molecules or crystals that are toxic to most immature mosquitoes. The various formulations of Bti the District uses contain no live bacteria but only the spores with protein molecules. Bti is fast acting and its efficacy can be evaluated almost immediately. It can kill mosquito larvae within 1 hour after ingestion, and since each instar must eat for the larvae to grow, Bti usually kills mosquito larvae within 48 hours of application. Bti leaves no residue and is quickly biodegraded. Bti efficacy is reduced in highly organic or polluted waters, low temperatures, areas with high larval densities, or when dense vegetative cover interferes with application at the mosquito-breeding site. Additionally, timing of the application is critical to maximize effectiveness, as the adult, pupal, and late 4th instar larval stages of mosquitoes are not susceptible to Bti.

Bti applied at label rates has virtually no adverse effects on applicators, livestock, or wildlife including beneficial insects, annelid worms, corals, flatworms, crustaceans, mollusks, fish, amphibians, protozoa, sponges, reptiles, birds, or mammals (Becker 1998; Boisvert and Boisvert 2000; Brown et al. 2002; Caquet et al. 2011; Davis and Peterson 2008; deBarjac and Sutherland 1980; Eder and Schonbrunner 2010; Garcia et al. 1981; Gharib and Hilsenhoff 1988; Holck and Meek 1987; Hurst et al. 2007; Jackson et al. 2002; Knepper and Walker 1989; Lagadic et al. 2014; Lawler et al. 2000; Leclair et al. 1988; Marten et al. 1993; Merritt et al. 1989; Miura et al. 1980; Molloy 1992; Molloy and Jamnback 1981; Mulla et al. 1983, 1982a; Negri et al. 2009; Ostman et al. 2008; Purcell 1981; Reish et al. 1985; Russell et al. 2009; Siegel and Shadduck 1990; Siegel et al. 1987; Sternberg et al. 2012; Tietze et al. 1993; Tozer and Garcia 1990; Waller

1992; Wipfli and Merritt 1994). However, nontarget activity on larvae of insect species normally associated with mosquito larvae in aquatic habitats has been observed. Impacts have been reported in some larvae belonging to the midge families Chironomidae, Ceratopogonidae, and Dixidae (Ali et al. 2008; Anderson et al. 1996; Liber et al. 1998; Lundstrom et al. 2009, 2010; Molloy 1992; Mulla et al. 1990a; Rodcharoen et al. 1991; Tozer and Garcia 1990; Vaughn et al. 2008). These nontarget insect species, taxonomically closely related to mosquitoes and black flies, apparently contain the necessary gut pH and enzymes to activate the Bti delta-endotoxins. However, the concentration of Bti required to cause these effects is many times higher than maximum allowed label rates for mosquito control.

Concerns have been expressed about the potential indirect effects that may exist for some nontarget organisms. Three studies have suggested possible food-web effects as a result of observed changes in invertebrate and vertebrate populations following use of Bti for mosquito control.

> Hershey et al. (1998) examined the effects of multiple spring and summer Bti and methoprene applications over a 3-year period (1991–1993) on the benthic macroinvertebrate communities of 27 wetland ecosystems in Wright County, Minnesota. Their study was part of a larger before and after study reported by Niemi et al. (1999). Reduced total insect density and richness were observed on Bti-treated sites for 1992 and 1993, with nematocerous diptera in the families Chironomidae, Ceratopogonidae, and Stratiomyidae the most affected. Minimal effects were observed on noninsect macroinvertebrates, and no negative effects on zooplankton or breeding birds were found as a result of treatments or changes to insect communities.

> Balcer et al. (1999) in a 1997–1998 follow-up study of these sites found no differences in the total mean density of insects and other invertebrates except for chironomid midges of the subfamily Chironomidae, which showed a reduction in density and biomass. They suggested that the drought conditions present during the earlier studies of Niemi et al. (1999) and Hershey et al. (1998), which would have increased susceptibility of organisms, may have contributed to the differences in the results observed. Additionally, Read (2002) noted that an analysis of the dosage data for these studies suggests that higher than planned doses in 1992 and 1993 may also have contributed to the difference in results observed.

> Poulin et al. (2010) reported reduced clutch size and fledgling survival of house martins in the Camargue region of France at Bti-treated sites. They found martin intake of nematocerous flies and their predators decreased significantly at Bti-treated sites and that consumption of smaller alternative prey including flying ants increased. They suggested that martin breeding success was affected by the decrease in nematocerous flies, specifically mosquitoes and certain Chironomid midges, which are a part of the martin diet. Their study did not, however, mention that the study area is an important rice-growing region nor did they assess the chemical usage on rice for pests. Furthermore, they did not discuss water management practices that directly affect aquatic insects.

A number of studies indicate use of Bti does not have indirect adverse impacts. Lagadic et al. (2014) reported the results of their 7-year study assessing the use of Bti on a 5-hectare tidal marsh at Locoal-Mendon, Morbihan, France. Their data indicated that long-term use of Bti had no influence on the temporal evolution of the taxonomic structure and abundance of nontarget invertebrate communities. They also found that the amount of invertebrates available as food resources for birds was maintained in Bti-treated sites. Two- and 5-year progress reports of this study, with detailed analysis of the time period covered, were also published by Caquet et al. (2011) and Roucaute et al. (2011). In each case the same conclusion, Bti-treated sites did not adversely impact nontarget aquatic invertebrate communities, was reached. Hanowski et al. (1997a,b) examined the effects of Bti treatments on birds in Wright County, Minnesota wetlands. They found no effects on the bird community or on 19 individual bird species. They also found no evidence that reproduction, growth, or foraging behavior of red-winged blackbirds was negatively impacted by the Bti treatments to the wetlands. Niemi et al. (1999) analyzes and discusses further the data collected and again concludes that although insect densities and biomass were reduced (mostly nematocerous flies), no negative effects on breeding birds could be attributed to the Bti treatments or changes to insect communities. Jackson et al. (2002) found that Bti applications to the Susquehanna River, Pennsylvania, did not significantly affect nontarget macroinvertebrates or fish. Fish species and composition did not change even though blackflies were a food source for some of the fish species present. Brancato (1996) reported no impacts to the fish community were found following eight Bti treatments of Aughwick Creek, Pennsylvania, to control black flies in 1995. Merritt et al. (1989) observed no effects to macroinvertebrate diversity or species richness following Bti treatment of the Betsie River in Benzie County, Michigan, for black flies. Overall, it would seem the potential indirect effects are therefore unlikely or minimal, making this material a useful, environmentally friendly tool for the management of mosquitoes.

The bacterium *Saccharopolyspora spinosa* produces compounds known as spinosyns, which effectively control all larval mosquitoes. The formulations registered and available for use contain spinosyn A and spinosyn D and do not contain any live bacteria. Factors that limit the effectiveness and use of this material include (1) sites with high organic content as the insecticide is readily absorbed and binds to particulate matter (Hertlein et al. 2010); (2) sites subject to full sunlight as the main degradative pathway is photolysis (the half-life in water exposed to summer sunlight is 1 to 2 days); (3) sites with high water flow, which results in excessive dilution and sublethal dosing of mosquito larvae; and (4) dense vegetative cover, which limits the ability of liquid and even sometimes granular formulations from reaching the water containing mosquito larvae.

Additionally, some concern is associated with exposure of bees to spinosyns. Therefore the selection of formulation (liquid or granular) as well as the timing of application when working in close proximity to beehives and areas with populations of bees is important. Studies have demonstrated that the dry residues of applied liquid formulations of spinosyns on plants has no effect on honeybees although bees belonging to the genera Bombus and Megachile exhibited some sensitivity (Biondi et.al. 2012; Mayes et al. 2003; Miles 2003; Morandin et. al. 2005). Additionally, the application of liquid formulations during those times when bee activity is low minimizes potential exposure and allows for the applied material to dry prior to bee activity (http://www.2ndchance.info/fleas-spinosadGarden.pdf).

Nontarget studies with other organisms have indicated some risk when using this material, especially at the higher label rates for mosquito control when applied to small contained wetland systems with limited or no water exchange or flows (Duchet et al. 2008, 2010; Jones and Ottea 2013; Lawler and Dritz 2013). Hertlein et al. (2010) reviews the published and unpublished literature concerning the use of spinosad (spinosyns of *Saccharopolyspora spinosa*) as a larval mosquito control agent, discussing in detail testing methodologies, LC values, formulation types, mosquito species susceptibility, effects on mosquitofish, and affects to nontarget aquatic organisms. Their nontarget review, although short due to limited research addressing use of spinosyns for mosquito control, reiterates the need for additional studies concerning the impacts of spinosad on nontarget organisms that share larval mosquito habitats. They conclude that "spinosad appears minimally disruptive to most nontarget species tested thus far when applied at or near its proposed field use rates." Their suggestion that more research is needed parallels Jones and Ottea (2013) and Lawler and Dritz (2013), who suggest that more research would help to confirm if lower rates could be used to effectively control mosquitoes and also minimize potential impacts to nontarget aquatic organisms. This suggestion is based on the fact that the sensitivity of immature mosquitoes to spinosyns is much higher than tested nontargets.

Additional hazard analyses of Bs, Bti, and spinosad are contained in the Ecological & Human Health Assessment Report (in Section 4.3 Mosquito Larvicides) prepared for the PEIR as Appendix B.

2.11.2 Examples of Tool Use

Pyrenone 25-5, Natular, and various formulations of Bti

2.11.3 Applicability to District IMMP

The District currently uses the pyrethrin product Pyrenone 25-5 which is synergized with piperonyl butoxide as a mosquito adulticide. The method of application is restricted to ULV fogging using truck-mounted, ATV-mounted, and handheld fogging machines. Most applications are associated with increased mosquito-borne disease (e.g. West Nile virus) risk, and typically occur in late summer and early fall. The District uses various formulations of Bti products (liquid, powder, granules, and water soluble packets) for management of immature mosquito populations to prevent adult emergence. *Saccharopolyspora spinosa* is currently only used in granular and tablet forms but may be used in liquid form in the future.

2.12 Insect Growth Regulators

2.12.1 Description

Insect Growth Regulators (IGRs) target immature insect populations. IGRs can be target specific, depending on the formulation used and the concentration that is applied to the target population of insects being managed. Therefore, adhering to label requirements and when used in the manner for which they are designed, IGRs affect the juvenile stages of the target organisms while causing little or no effects to the nontargets present (e.g., methoprene and mosquitoes). Unlike many traditional insecticides, IGRs do not affect an insect's nervous system, nor do they kill adult mosquitoes. Rather, IGRs prevent the ability of the immature stages to complete their final molt from the pupal stage to adult (prevent adult emergence).

The IGR currently used, and that has been used by the District for more than 2 decades, is s-methoprene. S-Methoprene (known simply as methoprene or as its trade name, Altosid) is a synthetic analogue (mimic) of a naturally occurring insect hormone called Juvenile Hormone (JH). JH is found during the mosquito's aquatic life stages and in other insects, but is most prevalent during the early instars. As mosquito larvae mature, the level of JH steadily declines until the 4^{th} instar molt, when levels are very low. This period when all the physical features of the adult begin to develop is considered sensitive. Methoprene in the aquatic habitat can be absorbed on contact and the immature mosquito's hormone system then becomes unbalanced. When happening during the sensitive period, the imbalance interferes with 4^{th} instar larval development. One effect is to prevent adults from emerging. Since pupae do not eat, they eventually deplete body stores of essential nutrients and then starve to death. Based on its mode of action, methoprene is considered an IGR. This material has no effect on mosquito pupae and must be contacted by larvae to be effective.

Methoprene is a material with very high specificity in its mode of action. Exhaustive reviews of the published literature on this material demonstrate that methoprene has little or no adverse environmental impact when used at label rates for mosquito control (Anderson et al. 1996; Butler et al. 2010; Glare and O'Callaghan 1999; Hanowski et al. 1997a,b; Henrick 2007; Kenyon and Kennedy 2001; Lawler et al. 2000; Mian and Mulla 1982; Office of the Minnesota Legislative Auditor 1999; Rexrode et al. 2008; Russell et al. 2009; Stark 2005). However, it has been suggested that potential direct and indirect effects may exist for some nontarget organisms subjected to repeated short-term or continuous long-term exposures of methoprene.

Most invertebrate field studies have shown minimal effects to nontarget organisms. Pinkney et al. (2000) investigated the repeated application of Altosid Liquid Larvicide to experimental ponds (rate of 3 ounces per acre or 0.16 ounce AI/acre) and found only isolated instances of reductions of aquatic nontargets. Overall, their analysis of the data indicated no significant differences between the Altosid and control ponds. Invertebrate populations in tidal salt marsh habitats treated with Bti, methoprene, or a combination of Bti and methoprene either were not affected or showed nominal effects with affected nontargets recovering quickly following exposure to methoprene (Lawler et al. 2000, Russell et al. 2009). Hershey et al. (1995) examined the effects of methoprene and Bti on nontarget insects in subdivided temporary woodland ponds and found no evidence of negative effects of larvicide treatments on density or biomass of any invertebrate group or in the richness of benthic fauna. Two studies, however, have suggested that repeated exposure and/or increased duration of exposure may increase the likelihood of nontarget indirect effects (e.g., reduced food resources). Hershey et al. (1998) performed a 3-year study evaluating insect populations in Wright County, Minnesota, wetlands that were treated 6 times from April through July of each year (1991–1993) with Bti and methoprene granules, and had a use pattern of methoprene that subjected wetland organisms to a continuous exposure during each year's test period. Methoprene had minimal effects on nontarget insect groups after the first year of treatment. Reductions in species richness, especially nematocerous flies in the families Tipulidae, Ceratopogonidae, and Chironomidae, were observed during the second and third year. Niemi et al. (1999) completed an integrated 6-year study that assessed the potential ecological impacts of Bti and methoprene use on zooplankton, insects, and breeding birds in Wright County, Minnesota wetlands. Their analysis included the data collected by Hershey et al. (1998), since the same wetlands and Hershey's work was a part of Niemi et al.'s original before and after study design. Changes in insect species richness reported by Niemi et al. was, therefore, simply additional analysis and discussion of the data already reported by Hershey. No negative effects were found to breeding birds or zooplankton as a result of exposure to methoprene, even though reductions in benthic insects were observed. Niemi et al. (1999) also noted that wetlands are ecologically complex and that other factors that may affect species distribution and abundance were not accounted for. Therefore, the lack of close coupling observed among birds, insects, and zooplankton suggests wetlands are highly complex ecological systems requiring additional study to better understand the many abiotic and biotic relationships that make up a wetland system.

Overall, methoprene is an effective tool for the management of immature mosquitoes when taking into account use patterns, material specificity (the large number of organisms that are unaffected at mosquito control rates), application methods, its rapid degradation in the environment, and the volume of published data indicating little or no adverse effects when used at mosquito control label rates. Additional hazard analysis of methoprene is provided in the Ecological & Human Health Assessment Report (see Section 4.3.4) prepared for the PEIR.

2.12.2 Examples of Tool Use

Methoprene liquid, pellets, granules, and briquettes used for control of mosquitoes in freshwater and tidal marshes, seasonal wetlands, ponds, fountains, water gardens, all types of man-made containers, septic tanks, wastewater ponds, winery waste ponds, etc.

2.12.3 Applicability to District IMMP

The District uses liquid, pellet, granular, and briquette formulations of S-methoprene products. Methoprene is a component of the District's IMMP and allows for control of mosquitoes by preventing adult emergence. The pellet and briquette formulations can provide control for several weeks, and all formulations are routinely rotated with the bacterial mosquito control products used by the District.

2.13 Mineral Oils/Surfactants

2.13.1 Description

Mineral oil and ethoxylated alcohol formulations (also known as surfactants) are used to control immature stages of mosquitoes (larvae and pupae). This control is accomplished by changing the surface tension of the water resulting in suffocation. These materials can also affect any adult mosquito that tries to land on the water to rest or lay eggs. Unfortunately, other air-breathing aquatic and semiaquatic insects including, but not limited to, water beetles, certain flies, water boatman, water striders and backswimmers, that are exposed to these surfactants can, while the surfactant is present, also be affected. Therefore, this tool is only used to prevent adult emergence when all other immature mosquito control methods have been deemed ineffective. The current surfactants available are BVA-2 Oil and Coco Bear Oil. The active ingredient in BVA-2 is mineral oil. Coco Bear Oil is comprised of 10 percent mineral oil with the remaining oil content consisting of food grade coconut and vegetable oils. An additional surfactant of interest but no longer available is Agnique MMF. This material is 100 percent ethoxylated alcohol.

The concept of using a surface agent to control immature mosquitoes is not new. One of the earliest accounts recommending this approach was described on August 29, 1793, in a Philadelphia newspaper known as *Dunlop's American Daily Advertiser*. The author, known only as A.B., described the use of "any common oil" to effectively control immature mosquitoes in rainwater barrels and cisterns. Interestingly enough, it would take approximately 100 years before this suggested management technique would be employed as part of organized mosquito control programs (A.B. 1793).

Early mosquito control programs generally used kerosene or coal oil whose use and effectiveness was well documented (Beutenmuller 1890; Cattell 1903; Chase and Nyhen 1902; Goldberger 1908; Gray 1903; Howard 1893, 1901, 1910, 1911; Howard et al. 1912; Mitchell 1907; Weed 1895). Unfortunately, these early materials were unsightly, had an odor, and could persist for some time at the application site. Research and development through the years has resulted in more effective formulations with better spreading capabilities, little or no odor, and very short-term persistence. Although surfactants are not the preferred method for managing immature mosquitoes, the new generation of these materials remain a part of organized mosquito control programs and are used only when necessary and appropriate, as part of the integrated management of mosquito populations.

Stage (1952) summarizes the history and use of petroleum oils for mosquito control. In his review he also notes those factors important for an oil to be effective for mosquito control purposes. These factors are (1) able to kill immature mosquitoes within a short period of time, (2) has a low surface tension, (3) is readily able to move around and through aquatic surface debris and emergent vegetation, (4) has a low toxicity to aquatic plants and wildlife, (5) is persistent only long enough to be effective, (6) is readily available, and (7) is low in cost. Finding a commercially available mosquito control surfactant that possesses all of these qualities has had its challenges and as such has resulted in a number of registered products that have come and gone during the last few decades (i.e., FLIT MLO, Golden Bear 1356, Golden Bear 1313, Golden Bear 1111). The two remaining surfactants available for mosquito control purposes, BVA-2 and CoCo Bear, are fairly new, function in a similar manner to the older products, and contain 97 and 10 percent mineral oil, respectively. Therefore, since an extremely limited body of literature is available concerning BVA-2 or CoCo Bear, literature for Flit MLO and Golden Bear products will be used.

Four studies by Tietze et al. (1991, 1992, 1993, 1994) tested three species of fish (inland silversides, mosquitofish, and sheepshead minnows) and a range of microorganisms and concluded that GB-1111 oil was not toxic to the tested organisms at label application rates. Mulla and Darwazeh (1981) experimented with GB-1111 in small experimental ponds and found that benthic invertebrates were unaffected while populations of surface-breathing insects were temporarily reduced following application of this larvicide. Miles et al. (2002) completed a significant independent study of nontarget effects of GB-1111, with US Fish and Wildlife Service's financial assistance, on the tidal marshes of Newark, California, and observed the following effects: (1) surface-breathing insect populations were reduced at the time of treatment; (2) this effect did not persist beyond a few days (= no residual pesticide effects); (3) those potentially affected invertebrates with high mobility left the site, while some of those that could not leave died (especially water boatmen [*Corixidae*]); and (4) overall populations of invertebrate species were not affected, apparently because of recolonization from neighboring untreated sites. They also examined the potential effects on mallard ducklings and noted that the ducklings showed no significant effects of weight loss due to depletion of invertebrate prey. It was observed, however, that some initial oiling and consequent matting of feathers occurred, but that survival, mass and weight gain, and overall condition remained good. Miles et al. (2001) also examined in a lab setting the effects of GB-1111 on hatching success of red-winged blackbird and bobwhite quail eggs and observed reduced hatching success when the eggs were treated with 3 and 10 times the maximum field application (label) rates. It is important to note though that at maximum label rates no significant effects to avian embryos were observed. Subsequent studies by Albers et al. (2003) and Hoffman et al. (2004) yielded similar results when eggs of mallard ducks, red-winged blackbird, and bobwhite quail were exposed to GB-1111 at 3 and 10 times maximum label rates. Both studies also concluded that the potential hazard to embryos was minimal until maximum label rates were exceeded. An earlier study by Albers and Heinz (1983) examined the hatchability of mallard duck eggs and duckling behavior following different levels of exposure to FLIT MLO. At 3 times the maximum label rate, egg hatchability was significantly reduced and changes in behavior, specifically reduced avoidance response, were observed. They concluded that application within label rates did not appear to pose a risk to the embryos of breeding birds.

2.13.2 Examples of Tool Use

BVA-2 Oil, CoCo Bear Oil.

2.13.3 Applicability to District IMMP

Although the District uses surfactants as part of its IMMP, the use is limited and closely monitored. Surfactants are a last line of defense when all other larval control measures have failed or have been deemed not possible. The District realizes very short-term effects can affect other air-breathing aquatic insects, such as diving beetles, backswimmers, water boatmen, and water striders. Some of these insects are also predators of immature mosquitoes. The District makes every effort to encourage and promote the presence of natural mosquito predators, which helps reduce the use of pesticides. The effects of surfactants are temporary, lasting until the dissipation of the material, which is typically within a couple of days. Furthermore, because District larvicide and pupacide surfactant use protocols require application of these materials only in areas with immature mosquitoes, and because immature mosquito distribution is highly patchy (Service 1993), recolonization of impacted nontargets from unsprayed areas should still occur promptly (Lawler et al, 1998; Miles et al, 2002; Mulla et al, 1981). Therefore, mineral oils/surfactants are a part of the District's existing IMMP. BVA-2 is currently used to manage pupal and late stage larval mosquitoes. CoCo Bear Oil is not used at this time but may be used in the future.

2.14 Mass Trapping

2.14.1 Description

The traditional approach to mosquito control where large areas need to be physically manipulated and/or treated with an insecticide can have many potential issues including, but not limited to, high cost, resistance of target mosquitoes, public acceptance, and risk of effects to sensitive habitats and nontarget species. These concerns coupled with the public's desire to reduce their interactions with mosquitoes continue to inspire research into new less toxic mosquito management strategies. Trapping has been an area of particular interest.

Many types of traps and trapping strategies are available for use. Mass trapping uses large numbers of traps, baited with a strong lure (e.g., carbon dioxide (CO_2), octenol, lactic acid, heat, certain wavelengths of light and sound, and food items such as sugars and proteins), which are placed in an effort to catch sufficient target pests to reduce the population to healthful levels. Depending on the species and density of the managed mosquito population, traps may be distributed over a large area. Lures for mosquitoes include, but are not limited to, CO_2, light, heat, and octenol. Yellow jacket traps use heptyl butyrate, sugars (e.g., fruits) and/or proteins (meats). An insecticide, rodenticide, food, or a sticky insert may also be used in the trap. Traps using a toxicant or electric grids are covered below in Section 2.15, Attract and Kill.

2.14.1.1 Mass Trapping Mosquitoes

The use of depletion or mass trapping as a possible alternative and/or supplement to the use of pesticides has received considerable attention (Adams 1996; Consumer Reports 2003; Day and Sjogren 1994; Henderson et al. 2006; Hougaard and Dickson 1999; Jackson et al, 2012; Kline 2002, 2006, 2007; Kline and Lemire 1998; Ogawa 1988; Quarles 2003; Smith et al. 2010; Weidhaas and Haile 1978). This technique uses specialized traps, which may also contain attractants to enhance their effectiveness, for collecting large numbers of mosquitoes. Recent advances in trap design and advances in understanding the biochemical cues and other factors that attract different mosquitoes to their potential hosts have begun to illustrate the possibilities as well as the limitations of mass trapping as a potential management tool.

For example, Revay et al. (2013b) tested the efficiency of seven commercially available mosquito host protection devices, including three spatial repellent-based products, one type of citronella candle, and three traps with CO_2 and/or ultraviolet light attractants. Their data indicated that the ThermaCELL Patio Lantern repelled the most mosquitoes at distances of 10 feet or more from the host when compared to controls. Mosquito traps with attractants on the other hand either increased or had no effect on mosquito biting activity at distances of 10 feet or less from the host when compared with unprotected controls. They also found that the placement of four of any one type of trap, one at each corner of a 4,050- and 8,192-square-meter area, yielded the best protection, with the Blue Rhino Trap being the most efficient, demonstrating a 40.1 and 18.1 percent reduction in biting activity, respectively.

Henderson et al. (2006) tested the effectiveness of the Mosquito Magnet Pro, which releases a combination of CO_2, heat, and moisture, to reduce mosquito abundance in both a rural and urban setting. Although six different species and nearly 2,000,000 mosquitoes were collected over a total of 94 trap nights, they found that continuous operation of these traps did not significantly reduce mosquito activity for either setting. Hougaard and Dickson (1999) tested both the ABC Pro and the Mosquito Magnet traps for their efficacy in managing adult western treehole mosquito (*Aedes sierrensis*) populations and found the Mosquito Magnet had trapped more treehole mosquitoes while the ABC Pro trap had collected more little house mosquitoes (*Culex pipiens*). They concluded from their data that the Mosquito Magnet was an effective tool in "helping control" western treehole mosquitoes in Salt Lake City, Utah. They also pointed out that neither trap eliminated all of the mosquitoes but they did help, especially with the homeowners who felt that some form of effort was being made to address a challenging mosquito problem where the effectiveness of traditional control methods was limited. Kline (2006) noted the results of two unpublished mass trapping experiments conducted in Florida between 2002 and 2004 using Mosquito Magnet Pro traps. The first was on three small islands associated with Lower Suwannee Wildlife Refuge where enormous populations of the salt marsh mosquito (*Ochlerotatus taeniorhynchus)* would make visitation impossible from May through October. One 23-acre island was selected and a minimum of 21 traps were used continuously from June to October of 2003 and 2004. Overall, a significant reduction in adult biting activity occurred such that no repellent was required while on the island. The second study was a collaborative effort with the US Department of Agriculture and involved a residential area in Gainesville. Two separate neighborhoods were surrounded by 12 Mosquito Magnet Pro traps that also used octenol as an additional attractant. Initial analysis of the data indicated a moderate level of control with a 50 percent reduction in mosquito levels in treated versus untreated residential neighborhoods. Smith et al. (2010) used 12 Mosquito Magnet-X traps at a coastal Florida state park and found that the traps did not significantly reduce mosquito numbers compared to the control sites. They further noted that during the latter part of the study mosquito numbers had reached such severe levels that park management requested spraying because of the number of complaints received from users of the park's facilities. They concluded that either more traps, a smaller treatment area, lower mosquito population levels, or some combination of all three would be necessary to achieve nonpesticide control using mass trapping.

Kline (2006, 2007) also provides an overview of the recent advancements in mass trapping technology and its potential as a mosquito management tool. He notes a number of important concerns significant to the effectiveness of mass trapping as a mosquito management strategy. They are (1) a thorough understanding of the target mosquitoes' behavior, biology, and ecology; (2) which attractants work best since an attractant for one species of mosquito can be ineffective for another; (3) reproductive or biotic capacity; (4) spatial distribution, which affects placement of the traps; (5) dispersal capacity, as a high dispersal rate, especially with species that travel long distances, poses challenges with managing localized populations and increases the risk of reinvasion from other sites; (6) density of the mosquito population, which can influence the number of traps required; (7) design of the trap and attractant delivery system since no one trap works best for the collection of all species of mosquitoes; and (8) the willingness of the local citizenry to tolerate a lower level of mosquito control in some circumstances and situations. Other factors, such as wind, temperature, humidity, density of vegetative cover, species of mosquitoes present, and time of year, also play a part in the effectiveness of these types of traps.

Other potential methods of mass trapping include the use of sound and ovitraps. Walker (1999) and Mankin (2012) both provide a review of the application of sound as a tool for managing insects. Unfortunately, the bulk of the research deals with insects other than mosquitoes. However, a few studies seem to be interested in collecting male mosquitoes to learn more about male mosquito ecology and mating competiveness, or to test responses of different species of males to sounds. Ikeshoji et al. (1985) reported that when a black cloth was attached to the base of a tripod and sound traps suspended at different heights above the tripod, that the mean male age and the insemination rate of females resting near the sampling site decreased compared to controls. They suggested that removal of sound sensitive males by this approach had the potential to be more efficient than other possible male manipulation mosquito control techniques although long-term and larger-scale trapping experiments were needed. Ogawa (1988) working with *Mansonia* spp mosquitoes in Malaysia reported that mass trapping of males by an attracting sound was promising. Note though that dry ice and the odor of guinea pigs was also used with the sound traps as they enhanced the attraction of mosquitoes near the traps, most of which were *Mansonia uniformis*. Stone et al. (2013) examined the factors affecting the responsiveness of male *Aedes aegypti* and *Aedes polynesiensis* mosquitoes to sounds and the use of sound traps in the field to better understand male ecology and mating competiveness. They found that age and mating status influenced the overall responsiveness to sound, while size did not. Traps modified with a device to produce a tone of 465 Hertz collected 76.2 and 49.7 percent of male *Aedes aegypti* in lab cages and greenhouse enclosures, respectively. Traps modified to emit a tone of 440 Hertz collected up to 50.8 percent of male *Aedes polynesiensis* in lab enclosures. In field settings, captures of *A. polynesiensis* was higher than *A. aegypti*, although the numbers of male *A*. *aegypti* captured were low in all field settings. Lastly, when sound-emitting traps were placed 16.5 meters from male mating swarms no significant difference in the male capture rate occurred between experimental and control traps. Mass trapping of mosquitoes with sound for mosquito control purposes would at this point in time appear to be ineffective.

Ovitraps help assess egg-laying female activity and are widely used as a part of mosquito surveillance and monitoring. These types of traps are specifically designed to attract and sample gravid female mosquitoes, either directly or by means of the eggs that are deposited within the trap. The design varies depending on the species of mosquito being sampled. For example, *Culex* spp mosquitoes are sampled with larger gravid traps such as the Reiter Gravid Trap. This device has a large dishpan-like plastic tub base, filled with hay-infused water or some other water with an attractant, and a small fan with a collection net or chamber placed above to suck in egg-laying mosquitoes that are attracted to the water. On the other hand, sampling for certain species of *Aedes* mosquitoes is best accomplished with small dark-colored cups or black-painted jars containing water and an egg-laying surface such as a hardboard paddle or coarse paper strip or paper ring near the water surface. These small style ovitraps are used for sampling *Aedes* spp eggs and do not collect the egg-laying adults. Irrespective of the type of ovitrap used, this tool is not effective at capturing large numbers of mosquitoes and also has other limitations. First, these traps either collect female mosquitoes that have already taken at least one blood meal or the eggs of blood-fed mosquitoes. This approach is counter to a mosquito control agency's purpose and the public desire of minimizing human-mosquito interactions. Second, the water in the larger traps tends to have a rather strong, unpleasant odor. Having large numbers of these traps about would result in complaints concerning the "unusual" smells in one's yard or neighborhood. Third, these traps tend to be effective at trapping the adults or collecting the eggs of certain species of mosquitoes, especially those that breed in specific types of container water (e.g., little house, banded foul water, fish pond, and some species of *Aedes* mosquitoes such as western treehole, Asian tiger, and yellow fever). Therefore, although useful for assessing female mosquito egg-laying activity, these traps do not appear to be a viable means for significantly reducing mosquito populations.

2.14.2 Examples of Tool Use

The District does not employ mass trapping as an abatement measure for the reduction of mosquito populations.

2.14.3 Applicability to IMMP

Operational difficulties exist in placing out and retrieving large numbers of traps for mosquitoes, the least of which are the volume of traps required, numbers of staff, amount of staff time, access, and travel necessary for this tool to be effective. Mass trapping of mosquitoes has proven to be both costly and in most instances ineffective. Therefore, the District does not use mass trapping as a tool for the abatement of mosquito populations. Instead, trapping is used to help assess mosquito and mosquito-borne disease presence and abundance and guide the application of other methodologies such as source reduction, sanitation, biological control, public education, and when necessary the use of pesticides.

2.15 Attract and Kill

2.15.1 Description

Attract and kill involves the use of a lure to draw the target mosquitoes to a location where the mosquito dies after either feeding on or crawling over the pesticide-lure mixture, crawling over or touching an electric grid, or being physically killed by some other mechanical means. Many different kinds of attract and kill devices exist, such as mechanical snap traps, bug zappers, sticky card traps, and various types of bait stations. Some are specific to mosquitoes (e.g., ATSB bait stations) while others may be more general in nature (e.g., bug zappers). Their placement and use vary and care must be taken when working with this methodology to optimize the intended result.

2.15.1.1 Bug Zapper/Electrocuters

Electric insect management devices or "bug zappers" have a long and varied history with the first publication of an electric fly control device appearing in a 1911 edition of *Popular Mechanics Magazine*. More efficient versions of electric insect control devices to help homeowners and farmers appeared in the same magazine in 1931 and 1934. The first patent was issued in 1934, although it would be a few years before the bug zapper would be readily available on the commercial market.

A bug zapper is a device that attracts and kills flying insects with an electric current. These devices typically consist of a protective cage of plastic or grounded metal bars that has inside an electrified metal grid with an internal fluorescent light source for emitting violet and ultraviolet light. The protective outer cage prevents people and animals (excluding insects) from touching the high-voltage grid. The light attracts insects to the metal grid and when they land on the grid they are electrocuted.

Unfortunately these traps are not effective at killing biting mosquitoes and instead kill large numbers of harmless and beneficial insects (Frick and Tallamy 1996; Lewis 1996; Nasci et al. 1983; Science Daily 1997; Surgeoner and Helson 1977). Frick and Tallamy (1996) assessed the insects that were killed in electric insect traps from 6 sites in Newark, Delaware. They noted that of the 13,789 total insects electrocuted, 18 or 0.13 percent were female mosquitoes. Additionally, 1,868 (13.5 percent) predatory and parasitic insects and 6,670 (48.4 percent) aquatic insects were killed. Their data suggested, albeit circumstantially, that the bug zapper was not an effective means for significantly reducing mosquitoes in one's yard. Nasci et al. (1983) tested the ability of these types of devices to reduce mosquito biting activity in 6 adjacent backyards in South Bend, Indiana. They observed that only 3.3 percent of the 3,212 insects killed on an average night were female mosquitoes. They also noted that humans were more attractive to mosquitoes than the bug zappers. Surgeoner and Helson (1977) evaluated the effectiveness of electric grid light traps in backyards in southern Ontario and concluded that they did not prove effective in reducing mosquito biting activity. When the release of CO_2 was added to the tops of the trap, their effectiveness significantly increased. Still they noted that these traps destroyed large numbers of other types of insects and that mosquitoes constituted a small portion of the total number of insects killed.

Another issue generally overlooked by the general user of these traps is the potential for release of air-borne insect particles and microbial contaminants. A number of studies examined the potential for this issue and noted that indeed a release of such contaminants occurred when insects, especially certain kinds of flies and moths, were disintegrated in electrocuting insect traps (Ananth et al. 1992; Broce 1993; Pickens 1989; Tesch and Goodman 1995; Urban and Broce 2000). Pickens (1989) further noted that trap design, placement height of the traps above the floor, and potential air movement or wind velocity were important factors in determining the distance of scatter of dismembered parts of electrocuted flies. Under normal circumstances and with proper trap placement, he suggested that a distance of 2 meters between wall-mounted traps and work areas would provide sufficient space to prevent potential contamination. Unfortunately it may not be the case for traps used in one's backyard, especially those hanging from porch, patio, or deck areas. Again trap design, placement, potential air currents and numbers of insects being zapped at any one time will be the determining factors concerning risks for potential contamination of food and drink items.

2.15.1.2 Attractive Toxic Sugar Baits (Mosquitoes)

Attractive toxic sugar baits (ATSBs) are a technology garnering considerable attention as a potential tool for the management of adult mosquito populations (Beier et al. 2012; Khallaayoune et al. 2013; Muller et al. 2008c, 2010a,b,c; Muller and Schlein 2008; Naranjo et al. 2013; Qualls et al. 2014, 2012; Revay et al. 2014; Xue et al. 2006, 2008, 2011). This strategy takes advantage of the fact that adult mosquitoes feed on plant sugars, which are an important source of energy influencing adult mosquito longevity, reproductive capacity, host seeking, and the potential to transmit disease (Andersson 1990; Andersson and Jaenson 1987; Magnarelli 1979, 1983; Nayar and Pierce. 1980; Nayar and Sauerman 1971; 1975; Yuval 1992). Both genders of mosquitoes feed on plant sugars, with some species exhibiting preferences for certain types of nectars or sugars (Grimstad and DeFoliart 1974: Muller et al. 2010a, 2011; Schlein and Muller 2008). Potential sugar sources can include, but are not limited to, flowers, sap, juices of decaying fruits, and honeydew.

ATSBs typically are solutions that consist of a sugar bait base, which contains a toxicant such as boric acid, dinotefuran, Saccharopolyspora spinosa, fipronil, eugenol, garlic oil, or some other microencapsulated insecticide or plant essential oil/extract. Adult mosquitoes are attracted to the ATSB, which may either be contained within a bait station with an accessible surface allowing mosquitoes to feed on the bait, or is applied as a liquid spray to the foliage of plants and man-made nonporous surfaces (e.g., painted or stained wood, metal, and plastic). Mosquitoes are then killed following ingestion of the ATSB.

Current research results indicate this approach has promise although with some concerns and limitations. First, a limited amount of data exists concerning impacts to nontarget organisms (Khallaayoune et al. 2013; Qualls et al. 2014; Revay et al. 2014). Although the potential impacts to nontarget organisms appear low, more research needs to be done to confirm these results with different formulations, application methods, and in different settings. Second, ATSBs require that a sufficient number of bait stations be placed within the area of mosquito activity (Xue et al. 2008) or that a certain amount of the surface area within a treatment site be covered with the ATSB solution to maximize effectiveness. Sprayed ATSBs must also be applied in a way that uniformly wets, and depending on the formulation, saturates the surfaces of treated foliage and nonporous surfaces. Third, although low, a risk exists that some surfaces may discolor when sprayed with certain liquid ATSB formulations (Universal Pest Solutions Terminix AllClear label 2013). Therefore, care must be taken to minimize potential discoloration of treated surfaces. Fourth, hand watering or automatic irrigation within sites that have been sprayed with an ATSB must be avoided for at least 24 hours, otherwise the material may be washed off and rendered ineffective.

2.15.2 Examples of Tool Use

Attractive toxic baits for the management of adult mosquitoes are a recently developed control strategy, with one product, Terminix® AllClear, commercially available.

2.15.3 Applicability to District IMMP

Due to the limited effectiveness of insect electrocution traps, and their propensity for destroying large numbers of nontarget insects, the District does not use these devices as a part of its IMMP. The District also does not recommend the use of bug zappers to its constituents as a method for dealing with the presence of biting mosquitoes.

ATSBs for the management of adult mosquitoes show promise but are limited in their availability. The District is aware of one commercially available product, Terminix® AllClear, which contains an essential oil of garlic. This product is new to the market, registered and released for use in California as of July 2014. The District still needs to operationally test this material, as well as other potential ATSBs, to determine those circumstances where their use may be effective while also having little or no nontarget species impacts. Therefore, although currently not used, the District reserves the right to use ATSBs in the future as a part of its IMMP (which may require additional CEQA analysis).

2.16 Inundative Releases

2.16.1 Description

Inundative releases are large-scale, periodic releases of parasites or predators to quickly reduce mosquito populations. This technique also includes the release of large numbers of genetically modified mosquitoes that have been irradiated, chemosterilized, or have had a gene altered. Inundative releases of predators and parasites can be used in situations where the existing levels of natural enemies are unable to sufficiently reduce mosquito populations to healthful levels. The use of genetically modified mosquitoes can be for population suppression or to reduce the ability of mosquitoes to harbor and transmit disease. The release of irradiated or chemosterilzed males is similar to the release of predators and parasites in that the goal is mosquito population reduction. Releases of mosquito natural enemies or sterile males is not self-sustaining and must be periodically repeated to provide effective long-term control. The use of gene-altered mosquitoes does not have to be regularly repeated as the goal is to introduce a gene into the mosquito population that is self-sustaining. This introduced gene changes the mosquito population to a less harmful form and/or reduces or eliminates the mosquito population entirely. Since potential and known predators and parasites of mosquitoes have been discussed above in Sections 2.5 through 2.9, they will not be addressed further here. Instead, this section will focus on the use of genetically modified mosquitoes. The following discussion is a brief summation of some of the literature concerning genetically modified mosquitoes and their future potential as part of an integrated mosquito management program:

2.16.1.1 Genetically Modified Mosquitoes

The use of sterilized or genetically altered mosquitoes for the management of mosquitoes and/or mosquito-borne disease has received and continues to receive considerable attention (Alphey et al. 2002, 2010, 2011; Bellini et al. 2013a,b; Benedict and Robinson 2003; Cha et al. 2006; Corby-Harris et al. 2010; Dame 1985; Dame et al. 1974, 2009; Gould et al. 2006; Harris et al. 2011; Ito et al. 2002; Knols et al. 2007; Laven 1967; Lavery et al. 2008; Lofgren, et al. 1974; Lowe, et al. 1974; Macer 2005; Morlan et al. 1962; Patterson et al. 1968, 1970, 1975, 1977; Reisen et al. 1982; Scott et al. 2002; Toure et al. 2004; Toure and Knols 2006; Weidhaas 1972; Weidhaas et al. 1962, 1974; Wise de Valdez et al. 2011). This interest has spanned more than 50 years and has intensified with advancements in technology; increased understanding of mosquito population biology, ecology and behavior; pesticide resistance; and malaria drug resistance. Areas of interest include, but are not limited to, male fitness or mating competitiveness, ecology and behavior of genetically modified mosquitoes, field performance, sterilization methods and materials, release methods, site assessment and selection, cultural issues and public concerns, ethical and legal issues concerning the use of genetically modified organisms, biosafety and risk assessment of using them, sustainability of introduced desired traits into a mosquito population, and effectiveness of reducing mosquito-borne disease transmission.

Success with the use of this approach has been inconsistent. Benedict and Robinson (2003) summarize the results of sterile and incompatible male releases (also known as sterile insect technique [SIT]) and note that regardless of mosquito species three significant factors have contributed to the observed field failures. The significant factors are production below desired levels, loss of male fitness, and immigration of mosquitoes into the release areas. Mosquito population levels and geographic distribution of the population to be treated may also contribute to the success of sterile male releases for the suppression of mosquito populations. For example, when working with isolated populations (e.g., populations within an isolated geographic area) and moderate population levels, SIT has been effective (Patterson et al. 1970; Weidhaas et al. 1974).

Whether or not SIT can work and be sustainable over a very large geographic area, as well as in circumstances with multiple species of mosquitoes, is not clear at this time. The release of sterile mosquitoes is a complex process involving initial colonization of the relevant species, mass rearing of competitive males for release, packing, transport, and release at the optimum place and time (Dame 1985). Having a good understanding of target population size, which helps determine the release period, number of releases, and ratio of sterile males to indigenous males released, is also important for successful use of this technique. Reduction of male competitiveness by radiation, immigration of fertilized females from outside release zones, and inability of laboratory-bred males to perform in the wild are some of the factors observed to affect SIT efficacy in some field tests (Dame et al. 2009). Even with significant advances in technology and understanding of mosquito population ecology, much is still to be learned about the application and effectiveness of SIT as a potential tool for integrated mosquito management.

Mosquitoes that have had their genetic makeup altered to reduce their ability to harbor and transmit mosquito-borne diseases such as malaria and dengue have also shown promise. This approach requires the use of a genetically engineered system to give the mosquitoes the desired trait as well as a system, known as "gene drive," to successfully spread the desirable gene into the wild mosquito population. Gene drive is important as it ensures that the desirable gene is passed on to more than half of the mosquito population, continues to spread, and will ultimately replace the undesirable trait (i.e., the ability to harbor and ultimately transmit a pathogen such as malaria). Gene replacement is different from more traditional SIT forms, which are self-limiting and usually emphasize population suppression rather than replacement, the release of large numbers of sterile or incompatible male individuals, and require repeated releases.

One of the more prominent issues associated with genetically modified mosquitoes is public perception. The release of genetically altered organisms into the environment, safety to humans, what they will do, what they may become, potential unforeseen effects, and the actual effectiveness to suppress mosquito populations and/or transmission of mosquito-borne diseases are all important questions being posed. A number of articles have been published discussing the moral, ethical, social, and legal issues concerning the use of genetically modified organisms for public health purposes (Alphey et al. 2010; Knols et al. 2007; Lavery et al. 2008; Macer 2005, 2007; Ostera and Gosten 2011; Toure and Knols 2006; Toure et al. 2004). Public concern about genetically modified organisms, some of which has been propagated by media sensationalism of "new or mutant organisms," is growing

Ecological and population biology issues create serious challenges to the application of genetically modified mosquitoes for disease control (Scott et al. 2002). Biosafety and concerns involving genetically modified mosquitoes are also significant issues (Ostera and Gosten 2011). Whether a self-limiting (e.g., population suppression or "no bite") or a longer-term self-sustaining approach (e.g., introduction of a desired gene) is used, the success of both bite and no-bite strategies for genetically modified mosquitoes depends on the ability of the altered mosquitoes to spread through the wild population. Although a number of recent developments have occurred, none of the genetically modified mosquito methods has been adequately field-tested and consistently demonstrated as operationally effective. The circumstances by which this technology may be used with local mosquito populations are also unclear and will require significant study to determine if and where this technology can be effectively used. Therefore, the District does not at this time use genetically modified mosquitoes as part of its IMMP.

2.16.2 Examples of Tool Use

Inundative releases of genetically modified mosquitoes is still experimental.

2.16.3 Applicability to District IMMP

Genetically modified mosquitoes are not commercially available and, therefore, are not used by the District at this time.

2.17 Regulatory Control

2.17.1 Description

Governments use regulatory control measures such as quarantines and hold notices to prevent the human-aided movement of pests and/or items likely to harbor the pest into their jurisdiction or the movement of pests from infested areas into uninfested areas within their jurisdiction. Operationally, this control can also involve state and local regulations for the creation and management of water impoundments, stormwater runoff, water quality, restoration and/or management of wetland habitats, weed control, and refuse management.

2.17.2 Examples of Tool Use

CDFA and CDC inspection of cargo containers, imported tires, and imported plants (e.g., lucky bamboo). Both the State of California and Alameda County have regulations for the maintenance of wastewater ponds, storage of tires, management of refuse, septic systems, etc. This tool is used on a limited basis in that the District makes every effort to coordinate with federal, state, and local regulatory agencies to stay informed of potential introductions of pests or pending/proposed regulatory actions.

2.17.3 Applicability to District IMMP

Regulatory actions help to prevent the human-aided movement of unwanted pests. The adoption of regulations is lengthy, time intensive, expensive and uncertain as to the regulatory outcome. They do not reduce pest numbers or the ability of the pest to spread or relocate on its own. Moreover, regulatory controls are dependent upon state and federal agencies to initiate and implement, and thus this approach cannot assure that any project objectives would be achieved. Additionally, regulatory actions have the potential to create as well as eliminate additional mosquito habitats. Any habitats created will require future surveillance and possible maintenance to minimize potential mosquito activity (e.g., above- and belowground stormwater detention basins, flood management projects, seasonal wetland habitats). Therefore, the District makes every effort to coordinate with those regulatory entities concerning potential introductions of pests or regulatory actions including, but not limited to, weed and refuse management, stormwater runoff requirements, water quality, and restoration or enhancement of wetland habitats.

2.18 Repellents

2.18.1 Description

The District classifies repellents into the broad categories of nonchemical and chemical. Nonchemical repellents are further subdivided into mechanical (e.g., fans and sound-producing devices) and nonmechanical (e.g., mosquito plants, eucalyptus trees, castor oil plants), while chemical repellents are further subdivided into natural and synthetic. Repellents are used to protect individuals from potential interactions with mosquitoes (e.g., being bitten). They do not kill the pest, nor do they reduce pest numbers. Rather, they force the pests into adjacent areas away from the treated areas or individuals. The different kinds of repellents have various operational issues and, therefore, varying levels of success when used.

2.18.1.1 Mechanical Repellents

Mechanical repellents include devices that produce high frequency or ultrasonic sounds and fans, which create strong air currents. The concept of using sound to attract, repel and even possibly destroy mosquitoes has been around for about 65 years (Kahn and Offenhauser 1949). A significant amount of research has since been conducted on the effectiveness of sound-repelling devices with the majority of the research demonstrating these devices either do not work or have such limited effectiveness as to render them useless for the control of mosquitoes (Ahmad et al, 2007; Andrade and Bueno 2001; Andrade and Cabrini 2010; Belton 1981; Cabrini and Andrade 2006; Dryden et al. 1989; Foster and Lutes 1985; Garcia et al. 1976; Gorham 1974; Helson and Wright 1977; Huang and Subramanyam 2006; Jensen et al. 2000; Koehler et al. 1986; Kutz 1974; Lewis et al. 1982; Revay et al, 2013a; Schreck et al. 1977, 1984; Schreiber et al. 1991; Singleton 1977; Snow 1977; Yturralde and Hofstetter 2012). Coro and Suarez (1998, 2000) and Enayati et al. (2010) provide a more thorough review and history of sound-emitting mosquito-repelling devices reiterating the ineffectiveness of these devices. Coro and Suarez (2000) also express concern about the potential for human health issues as a result of exposure to the high intensity ultrasonic frequencies emitted by these devices.

The use of fans to repel mosquitoes has received recent attention in the popular media as an alternative to the use of chemical repellents and insecticidal sprays (Broad 2013; O'Connor 2010; Sagon 2013). Much of this attention stems from anecdotal reports of effectiveness and the interpretation of a study conducted by Hoffman and Miller (2003). Hoffman and Miller (2003) examined the effects of wind created by electric fans on the numbers of mosquitoes captured in CO_2-baited CDC light traps and found that the number of adults captured decreased with increasing wind velocity. They further found that by tripling the amount of CO_2 released they could double the number of mosquitoes captured, and from these data suggested that wind diminished mosquito catches mostly by diluting the presence of mosquito attractants rather than by exceeding mosquito flight capability. They recommended that wind generated from the use of fans should be researched further as a means of protecting people and their pets from mosquitoes in their backyards. The idea of using fans to repel mosquitoes is interesting but has some inherent issues: (1) the number and placement of the fans needed to be effective; (2) the additional power usage as a result of having large numbers of citizens and small businesses all using fans to repel mosquitoes; (3) the limited area of protection provided, meaning when people leave the area where fans are creating currents the potential exists to be attacked by waiting mosquitoes; (4) some species of mosquitoes, marsh *Aedes* spp in particular, are quite capable of flying into strong currents to reach a host to obtain a blood meal; and (5) the number of potential hosts present in the area that increases the volume of mosquito attractants emitted (e.g., a dinner or garden party with multiple guests, or an outdoor business luncheon/party). These issues are but a few for which additional research is needed to better understand the feasibility of fan-generated wind currents as a potential repellent of mosquitoes.

2.18.1.2 Mosquito-Repelling Plants

The suggestion that the cultivation of certain plants around one's home or property could repel mosquitoes dates back more than 120 years (Riley and Howard 1893, 1894; Sanders 1893). At the time it was reported that the planting of stands of eucalyptus trees discouraged the presence of mosquitoes. Since then numerous anecdotal stories have come forward about other species of plants that purportedly had adult mosquito-repelling properties (e.g., castor oil [*Ricinus communis]*, basil [*Ocimum* spp], chinaberry tree [*Melia azedarach]*, and mosquito plants [*Pelargonium citrosum]*). The most famous of these plants is the mosquito, citrosa, or citronella plant, which received a significant amount of attention in the popular press in the late 1980s and early 1990s. Unfortunately, to date all potential "mosquito-repelling plants" have turned out to have little or no actual repelling capabilities when simply used as cultivated or potted plants (Cilek and Schreiber 1994; Cummings 1993; Cummings and Craig 1995; Howard 1901, 1910; Howard et al. 1912; Jensen et al. 2000; Lewis 1993; Matsuda et al. 1996; Quayle 1906). On the other hand, the essential oils derived from some of these and other plants have shown some promise as repellents when applied topically.

2.18.1.3 Chemical Repellents (Natural and Synthetic)

A fairly extensive volume of literature exists concerning the development, use, and health implications of chemical repellents for mosquitoes. One of the earliest reports involves the use of a mixture of mutton tallow and kerosene, which was then spread all over the burros of miners working in Minaret Mining District, California, to reduce the attacks of abundant mosquitoes and horse flies (Riley and Howard 1894). References to other early practices for human protection include (1) the use of essential oils such as citronella, camphor, cassia, peppermint, lavender, absinthium, American pennyroyal, and eucalyptus; (2) the use of materials such as lemon juice, vinegar, paraffin oil, and kerosene; and (3) and Native American techniques including the use of golden seal mixed with bear fat, western yarrow burned as a smudge, and smears of rancid alligator fat or mud (Beutenmuller 1890; Howard 1901, 1910, 1911; Howard et al. 1912; Giles 1900; Pierce 1918; Turner 2013).

Chemical repellents can be loosely grouped as natural and synthetic. Natural repellents include essential oils and those materials of plants and animals used as smudges and topical smears. Synthetic repellents are chemically created materials, which are typically used as topical or spatial agents to repel mosquitoes. Prior to 1945, the active ingredients available for use in insect repellents were primarily natural plant oils (e.g., citronella, camphor), pyrethrum (used as a powder and in coils), and three synthetics commonly known as DMP, Indalone, and Rutgers 612. The synthetic repellent (N,N-diethyl-m-toluamide) (DEET) was developed by the US Army in the 1940s, available for use by military personnel in 1946, and commercially available in 1957 (Debboun et al. 2007; EPA 1998, 2012; Jackson et al. 2008; NPIC 2008). The literature indicates a number of promising natural and synthetic repellents, although few have been rigorously tested for their effectiveness and safety, and fewer still have become commercially available.

Essential oils and plant extracts continue to receive significant attention for their potential as mosquito repellents. Amer and Mehlhorn (2006) used human volunteers to test the repellency effect of 41 essential oils against *Aedes aegypti*, *Anopheles stephensi,* and *Culex quinquefasciatus*. They found *Culex quinquefasciatus* was sensitive to all oils, *Anopheles stephensi* was moderately repelled, and *Aedes aegypti* was least repelled. The five best essential oils were litsea, cajeput, niaouli, violet, and catnip. They also reported that to achieve an 8-hour protection time and 100 percent repellency with the 5 best oils required the addition of 20 percent genapol and 10 percent polyethylene glycol to fix the aromatic components of the essential oils onto the skin. Numerous other studies have examined pine, peppermint, spearmint, geranium, clove, thyme, rosemary, black pepper, cardamom, cinnamon, coriander, sage, fennel, garlic, ginger, cedarwood, lavender, lemon grass, eucalyptus, basil, neem, and numerous other plant oils and extracts for their potential repellent properties (Ansari et al. 2000, 2005; Barnard 1999; Carroll and Loye 2006; Choi et al. 2002; Das, et al. 2003; Dekker et al. 2011; Hao et al. 2008, 2013; Kang et al. 2009; Kant and Bhatt 1994; Kim et al. 2005, 2012; Muller et al. 2009; Park et al. 2005; Phasomkusolsil and Soonwera 2010; Sharma, et al. 1993; Shooshtari et al. 2013; Tawatsin et al., 2001; Vongsombath et al., 2012; Webb and Russell 2007; Zhu et al. 2006). The results, although promising for some plant-based materials, vary widely with some achieving the level and length of protection that has been observed with the synthetic repellent DEET.

Maia and Moore (2011) provide a thorough review of plant-based repellents noting that commercial repellents containing plant-based products have increased in popularity with the general public. They further point out the popular misconception that repellents containing plant-based materials are safer than synthetic repellents and suggest a need for further standardized studies as few studies have followed the

World Health Organization's Pesticide Evaluation Scheme guidelines for repellent testing. Control and eradication of mosquitoes has become an important public health issue to reduce the many mosquito-borne illnesses. Urban expansion, deforestation, and industrialized farming have also increased the frequency of human-mosquito interactions. The need for more effective, environmentally friendly methods for managing mosquitoes and mosquito-borne disease has prompted additional research concerning repellents. Although some chemical mosquito repellents have been shown to be safe to humans, some concern remains about potential dermal and neurotoxicity (Patel et al. 2012). For example, Bakkali et al. (2008) showed that essential oils can act as prooxidants, and depending on concentration, can exhibit cytotoxic effects on living cells. Dweck (2009) reviews the toxicology of essential oils, discussing oils that may be skin irritants, phototoxic, carcinogenic, and cause potential pregnancy issues for women. He further states that just because a material is natural does not mean it is safe. Tisserand (2007) discusses the challenges of using essential oils in aromatherapy and states that proof of absolute safety does not exist. He also reviews toxicology and includes in his discussion constituents of essential oils with anticarcinogenic effects, balancing risk with benefit, and issues associated with misinformation and bias. Vigan (2010) discusses the renewed interest in essential oils, including definition, history, toxicity, uses, and European regulation that is being developed. Vigan also reiterates that just because essential oils are natural substances does not mean they are harmless.

The body of literature concerning synthetic repellents is also extensive although the focus has been more on toxicology and the development of spatial repellent systems rather than the identification of new synthetic compounds with repellent properties. Three materials, DEET, picaridin, and metofluthrin, are commercially available, with picaridin and metofluthrin having come to the US market within the last 10 years. DEET and Picaridin are topical repellents that can be applied to the skin while metofluthrin is a spatial repellent that cannot be used topically.

DEET is the most widely used insect repellent in the world. This repellent has demonstrated a broad range of effectiveness against mosquitoes, ticks, fleas, chiggers, and many other species of biting insects (Clopton and Gold 1992; Frances 1987; Frances et al. 1996; Gilbert et al. 1955; Mehr et al. 1984; Mount and Snoddy 1983; Qiu et al. 1998; Robert et al. 1992; Rutledge et al. 1978; Schreck et al. 1979, 1986). Although DEET is highly effective and of low toxicity, reports of adverse reactions range from skin sensitivity and rashes to rare instances of encephalopathy, seizures and death (Briassoulis et al. 2001; Clem et al. 1993; Koren et al. 2003; Maibach and Johnson 1975; Miller 1982; Reuveni and Yagupsky 1982; Roland et al. 1985; Shutty et al. 2013; Wantke et al, 1996). Osimitz and Grothaus (1995) assessed the safety of DEET, reviewing animal safety studies, case reports of human reactions, and literature reviews of poison control center data, and concluded that serious medical effects from the proper use of DEET was very low. They also stated that individuals who experienced serious side effects used DEET products with concentrations of 11 to 50 percent. Osimitz and Murphy (1997) performed a similar assessment addressing neurological effects only and concluded the risk of adverse symptoms appeared low when adhering to label directions. McGready et al. (2001) studied the effects of DEET in pregnant women and found that DEET was safe when used in the second and third trimesters of pregnancy. They further found no adverse effects on survival, growth, or development of the child at birth, or at 1 year post-birth. They concluded that the risk of DEET accumulating in the fetus was low and that DEET was safe to use in the last 2 trimesters of pregnancy.

The environmental impacts of DEET use are not well known, Costanzo et al. (2007) point out that DEET has been detected in water samples from many locations around the world. They also note that DEET is mobile and persistent but state that current information on DEET in the environment suggests the risk to aquatic organisms at current observed environmental concentrations is small. Lastly, they note that a full risk assessment was not possible as important data gaps existed, especially on transport, fate, and ecotoxicity.

Picaridin was developed in the 1980s but did not become commercially available within the US until 2005. This repellent is similar to the natural compound piperine, which is found in plants that are used to make the spice black pepper. Like DEET, picaridin is an effective repellent of many species of mosquitoes, biting flies, ticks, fleas, and chiggers. The toxicity classification is also similar to DEET with the exception that DEET has a higher primary skin irritation toxicity. Gervais et al. (2009) provides a good summary of the chemical properties of picaridin and known toxicological data pointing out existing data gaps.

Reports of adverse reactions to picaridin use are sparse. Corazza et al. (2005) report a case of contact dermatitis in a 39-year-old male that used a spray formulation containing 10 percent picaridin. Testing confirmed the patient's sensitivity to both the inactive ingredient, methyl glucose dioleate, and picaridin. Shutty et al. (2013) report the strong allergic response of a 22-year-old male to DEET and lack of response to picaridin. They concluded that open patch testing for contact allergy may be a helpful tool for helping individuals that suffer from allergic skin responses to insect repellents. They also suggested that patients sensitive to DEET may be tolerant of other insect repellents and that picaridin may be an acceptable alternative.

Metofluthrin is a synthetic pyrethroid that can be used as an insect repellent. Although capable of killing biting flies, metofluthrin has also been shown to be a spatial repellent of mosquitoes (Kawada et al. 2004a,b, 2005; Lucas et al. 2005, 2007; Xiu et al. 2012). However, some uncertainty seems to exist as to whether or not metofluthrin is actually a repellent or serves more as an insecticide because of its ability to effectively kill adult mosquitoes. Rapley et al. (2009) tested the effects of sustained release metofluthrin on mosquito biting and activity in an apartment setting and found that mosquitoes were not repelled from a room exposed to metofluthrin. Instead, many dead mosquitoes were found in the treated room. Furthermore, an adjacent room had greater than one-third mosquito mortality from spillover exposure to metofluthrin. Ritchie and Devine (2013) performed a similar study and found biting activity was significantly reduced as a result of the 80 to 90 percent mortality that occurred within the first hour of metofluthrin exposure. They also reported that female mosquitoes became disoriented, stopped landing on hosts, and sought out resting sites within the first few minutes. They concluded that metofluthrin did not result in mosquitoes leaving treated areas. These data suggest that determination of the concentration required for repellency rather than knockdown undoubtedly will vary depending on whether a person is outdoors or within an enclosed space such as a room.

It is not clear if metofluthrin affects a wide range of vectors like picaridin and DEET as few studies have been performed to determine the effectiveness of this material as a repellent against other vectors. Zollner and Orshan (2011) tested a fan vaporizer releasing metafluthrin in a field environment and found phlebotomine sand flies were not repelled. Published data concerning other vectors appear to be lacking.

2.18.1.4 Spatial Repellents/Repellent Systems

Spatial repellents are any chemical, which in the vapor phase affects biting insects at a distance from the release site and also inhibits vector host seeking and biting activity. Typical spatial repellent chemicals include both natural and synthetic materials such as essential oils, pyrethrins, allethrin, and metafluthrin. Although meant primarily to repel mosquitoes, some spatial repellents can also effectively knockdown or kill mosquitoes. Delivery methods of spatial repellents can be classified by the methods of vaporization, which are (1) heat-based vaporization (e.g., coils, candles, torches, and lamps with butane heat), (2) fan dispersed (stationary emitters and wearable personal devices), and (3) passive vaporization (impregnated paper and plastic strips). The effectiveness of spatial repellents depends on a number of factors including, but not limited to, the material used, effective distance from point of release (spatial action), persistence, the delivery method, the size of the area to be protected from mosquitoes, prevailing weather conditions, and species of mosquitoes present. Like any mosquito control material and management technique, selection of the proper material and delivery method is critical. Although useful, spatial repellents are typically meant to protect small localized areas (e.g., a room, patio area, or within a few feet around an individual). The concurrent use of multiple delivery systems (e.g., candles, coils, torches) can expand the protected area but also requires vigilance and proper maintenance to remain effective.

Repellent candles, diffusers, and torches have demonstrated variable levels of effectiveness depending on the type of essential oils used and mode of release. Lindsay et al. (1996) evaluated the effectiveness of 3 percent citronella candles and 5 percent citronella incense to repel field populations of *Aedes* spp mosquitoes. Overall, they found a 42.3 percent reduction in the numbers of bites received with citronella candles and a 24.2 percent reduction with citronella incense. Muller et al. (2008a) assessed the effectiveness of 5 percent citronella, linalool, and geraniol candles at repelling mosquitoes and sand flies and found geraniol candles were almost 5 times more effective than those containing citronella. Observed mosquito repellency rates were 29.0, 71.1, and 85.4 percent for citronella, linalool, and geraniol candles, respectively. Muller et al. (2008b) evaluated the same 3 types of essential oil candles in an outdoor setting and observed that the geraniol candle was again the most effective, reducing mosquito biting by 81.5 percent at a distance of up to 1 meter. They also reported that effective repellency dropped significantly as the distance from the candle was increased to 2 and 3 meters. They also assessed the ability of these same candles to protect volunteers in a high-biting-pressure environment and found that mosquito biting activity was reduced the most with geraniol candles by an average of 56 percent at distances of no more than 1 meter. Muller et al. (2009) assessed the degree of protection provided from mosquitoes by commercially available citronella, linalool, and geraniol candles and diffusers. They found that diffusers performed significantly better than candles in both indoor and outdoor environments and also had a greater effective distance (22, 58, and 75 percent repellency at 6 meters outdoors for citronella, linalool, and geraniol, respectively). Revay et al. (2012) tested the effectiveness of an area diffuser that released 0.3 percent aerosolized geraniol at timed intervals and claimed to provide protection up to a maximum distance of 18 feet. They found mosquito biting activity was significantly reduced with optimum protection afforded to volunteers that were within 9 feet of the diffuser and timed release intervals of 5 minutes. Revay et al. (2013b) evaluated the effectiveness of 7 commercial personal antimosquito protection products including 2 diffusers. One diffuser used 31.2 percent metafluthrin while the other used a mixture of essential oils (cinnamon 10.5, eugenol 13, geranium 21, peppermint 5.3, and lemongrass 2.6 percent). Both devices were found to be highly effective reducing mosquito biting from 87.55 to 97.22 percent, depending on the device and the species of mosquito tested.

Mosquito coils and incense have been around for more than 100 years and are one of the most widely used forms of nontopical repellents (Debboun et al. 2007). These devices generally consist of a combination of combustible active and inert ingredients (sawdust, a binder such as starch gel, a dye or colorant for appearance, a combustion regulator such as potassium nitrate, a perfume to make the smoke from the coil more acceptable, and either pyrethrum or a synthetic pyrethroid such as allethrin). Studies indicate an average of 80 percent effectiveness for a period of up to 8 hours under calm conditions (Strickman et al. 2009). Factors affecting effectiveness of coils varies with wind or air movement, spatial action, placement, and size of the area to be protected the most significant.

Although useful, concerns have been expressed about the potential health risks from smoke and by-products released when coils are used. Liu et al. (2003) examined the emissions from six different brands of coils used in China and Malaysia and found that pollutant concentrations could significantly exceed air quality standards and guidelines. They also found coil smoke contained a number of volatile organic compounds including three polycyclic aromatic hydrocarbons, benzo[a]pyrene, benzo[b]flouranthene and benzo[k]flouranthene, which have been classified by the US Environmental Protection Agency as probable human carcinogens. Lastly, formaldehyde emissions released from the burning of one coil was found to vary but could be as high as from the burning of 51 cigarettes. Therefore, they suggested that exposure to mosquito coil smoke posed significant acute and chronic health risks. Less specific were the epidemiological studies of Chen et al. (2008) and Koo and Ho (1994). Chen et al. (2008) administered questionnaires to 147 lung cancer patients and 400 potential controls gathering a broad range of data including the use of incense and mosquito coils. They found exposure to mosquito coil smoke was twice as high in cancer patients than controls. They suggested that the higher frequency of mosquito coil use increased the risk of lung cancer and that exposure to mosquito coil smoke further increased the risk of lung cancer in cigarette smokers. Chen et al. (2008) also reported that those who infrequently used mosquito coils also had a significantly higher risk of lung cancer than nonusers. Koo and Ho (1994) summarized the results of three Hong Kong epidemiological studies they performed between 1981 and 1988. They reported that analysis of air-borne particulates in homes indicated increases in the presence of benz[a]anthracene and benzo[k]flouranthene, both of which are considered carcinogenic compounds. They also reported chronic sputum rates for exposed children were twice as high as rates for unexposed children. Overall, they concluded that mosquito coil smoke is a source of air pollution that may cause increased bronchial irritation but that past coil smoke exposures was not found to be a risk factor for lung cancer.

Studies with lab rats have also indicated the potential for health risks associated with exposure to mosquito coil smoke (Garba et al. 2007; Liu and Sun 1988; Liu and Wong 1987; Okine et al. 2004). Effects to liver, lung, and spleen tissue as well as blood and the immune system were observed. Although these studies address potential affects from use of coils in indoor situations with minimal air circulation, outdoor exposure levels and risks are not adequately addressed.

2.18.1.5 Common Beliefs, Myths, and Home Remedies

A number of common beliefs exist about ways of reducing human-mosquito interactions without using traditional measures such as pesticides. Some of the more popular examples are consumption of garlic, beer, and vitamin B supplements, which are thought to function as systemic repellents. Unfortunately, none of these approaches has proved as effective as has been claimed.

Rajan et al. (2005) investigated the hypothesis that ingestion of garlic repels mosquitoes and found garlic provided no significant systemic repellence. They did, however, suggest that additional studies be performed to address whether continuous long-term consumption might yield different results. Maia and Moore (2011) reviewed the development, testing, and efficacy of plant-based insect repellents and reiterated that "the consumption of garlic has not been shown to be effective at repelling mosquitoes." Stjernberg and Berglund (2000) performed a study that examined the daily consumption of 1,200 mg of garlic to repel ticks. They concluded that garlic may be considered as an alternative to more traditional materials, which could have more adverse effects to the user. Unfortunately, the data presented did not really support such a claim as it was at best marginally better than using no repellent at all. This study also did not compare the effectiveness and safety of garlic to other commonly available repellents such as DEET or permethrin. Lastly, the World Health Organization’s Pesticide Evaluation Scheme Guidelines for repellent testing were not used making the data more anecdotal in nature.

The consumption of beer also has its issues although no doubt a person might be less likely to notice the effects of biting mosquitoes when their consumption has unfortunately been in excess. The hypothesis is beer contains vitamin B and other constituents that when consumed keep mosquitoes away from the person who drank the beer. Shirai et al. (2002) studied the effects of alcoholic consumption on mosquito biting activity. They found that mosquito landing activity was significantly higher after individuals ingested 12 fluid ounces (one can or bottle) of beer (ethanol concentration 5.5 percent) and concluded that humans are more attractive to mosquitoes after consumption of alcohol. Bernier et al. (2007) also report that in one of their studies, a test subject who regularly consumed alcohol, was the most attractive to *Aedes aegypti* mosquitoes.

Another home remedy is the consumption of vitamin B_1, B_2 and/or B_{12}, which is believed to be an effective systemic mosquito repellent. Shannon (1943) reported on the effects of administering up to 80 mg of thiamine chloride (vitamin B_1) per day to his patients that were "unusually attractive" to mosquitoes and also had significant reactions to the bites. He noted that vitamin B_1 reduced the numbers of bites received and the severity of the bite reaction. Unfortunately, his limited observations and lack of knowledge concerning the many other factors that may make a person unattractive to mosquitoes led him to claim that the ingestion of thiamine hydrochloride would effectively repel mosquitoes. Subsequent research has shown that ingestion of vitamin B complex supplements does not reduce a person's attractiveness to mosquitoes or the severity of their reactions to the bites. Ives et al. (2005) examined whether the ingestion of vitamin B supplements would repel the malaria mosquito *Anopheles stephensi* and observed that vitamin B supplements did not significantly reduce mosquito landing or biting activity on humans, Wilson et al. (1944) performed preliminary studies observing the systemic repellent effects of ingesting up to 505 milligrams of vitamin B_1. They found that *Aedes aegypti* mosquitoes were not repelled and that the biting rate and the human host's reactions to bites were no different than controls. Khan et al. (1969) also examined thiamine chloride as a potential systemic mosquito repellent. They found no significant differences in the attraction, probing time, or biting of *Aedes aegypti* mosquitoes between controls and test subjects. They also applied thiamine chloride as a topical agent and found no reduction in itching or wheal formation following mosquito bites and, therefore, concluded that vitamin B_1 was ineffective as a systemic mosquito repellent in man. Revay et al. (2013a) investigated the use of a transdermal patch containing 300 mg of vitamin B_1 and found that the patch did not provide significant protection from mosquito biting compared to volunteers that were not wearing the patch.

2.18.2 Examples of Tool Use

DEET, Picaridin. District staff may choose to use topical repellents and wear repellent treated clothing as a safety measure when performing their work for the District. The District, however, does not require that staff use repellents.

2.18.3 Applicability to District's IMMP

Repellents are not used as a control measure, since they will not reduce large numbers of problem insects or mammals and may even enlarge the infested area by driving the mosquitoes away from breeding and/or treated sites (Maia et al. 2012; Moore et al. 2007). Repellents also require proper application, timely use, and discipline concerning their use to achieve optimal effectiveness. Unfortunately, the use of repellents does not guarantee the elimination of human-mosquito interactions and potential mosquito-borne disease transmission.

It is important to note that repellents have a number of limiting factors: (1) persistence; (2) effectiveness over a wide range of mosquito species; (3) influence of climactic factors, which can limit distance, area, and/or period of effectiveness; (4) the sensitivity of the protected person or animal to the repellent being used (allergy and/or toxicity); (5) long-term effects due to prolonged use and repeated applications; and (6) effects to nontarget organisms when the repellent may get into unintended habitats or sources. Repellents can be helpful in reducing human-mosquito interactions but requires proper use and an understanding of the formulation used, species of mosquito(s) present, climactic factors at the time of use, and type of activity the protected individual is undertaking. Regardless of the type of chemical repellent used, it is important to carefully monitor proper use and the need for continued usage for a repellent to be optimally effective.

Additionally, because of the potential, albeit rare, possibility for adverse reactions when using some repellents, the District does not make recommendations to the public about the use of repellents and/or repellent systems. The District does, however, make available any information that it has on repellents to any person that asks and suggests they confer with the manufacturer and, when applicable, a licensed medical professional, prior to use.

Therefore, repellents are not a part of the District's IMMP.

3 Screening of Tools

Reasonable alternatives are developed through a review of the feasibility of all identified potential tools. To be feasible, an alternative should be capable of accomplishing project purposes in a successful manner in California within a reasonable period of time. This section explains the process for determining the components of the 2014 Program.

3.1 Program Objectives

The District undertakes activities through its Program to control and/or provide surveillance and information on mosquitoes, vectors of disease and/ or discomfort in the Program Area.

The Proposed Program's specific objectives are as follows:

> Reduce the potential for human and animal disease caused by mosquitoes

> Reduce the potential for human and animal discomfort or injury from mosquitoes

> Accomplish proactive, effective and environmentally sound mosquito management by means of:

  - Surveying for mosquito abundance/human contact
  - Establishing treatment criteria
  - Appropriately selecting from a wide range of Program tools or components

Most of the relevant mosquito species are quite mobile and cause the greatest hazard or discomfort at a distance from where they breed. Each mosquito species has a unique life cycle, and most of them occupy several types of habitats. To effectively control them, a proactive integrated mosquito management program must be employed. District policy is to identify those species that are currently vectors, to recommend techniques for their prevention and control, and to anticipate and minimize any new interactions between mosquitoes and humans. Furthermore, the District is committed to using the least environmentally disruptive tools in its IMMP. This commitment includes, but is not limited to, the following: continuous training of all staff on sensitive habitats, special status species, and mosquito biology and behavior; coordinating District activities with landowners, managers and resource agencies; participating in habitat restoration and enhancement projects; finding, researching and implementing new materials and technologies that help conserve environmental resources while also meeting the District's responsibility of protecting public health; and serving as a resource for information on mosquitoes, mosquito management strategies, sensitive habitats and special status species/species of concern.

There are circumstances when the District may not be able to effectively manage mosquito populations and meet all of the objectives above. Constraints such as access, weather, multiple sites producing mosquitoes at the same time, and acquisition of required permits can all affect the District's ability to optimally apply time sensitive materials and/or use the least environmentally disruptive tools. While these are the most frequently encountered and obvious limitations, another increasingly more significant constraint is working within the confines of conflicting local, state and federal ideologies and laws.

The District has long recognized the importance and value of coordinating and integrating its activities with the public, landowners, and the many resource agencies that operate within its Program Area. To narrowly interpret and then aggressively enforce those sections of the health and safety code that give the District its authority to manage mosquito populations ultimately eliminates effective communication and cooperation with the very people the District serves. Poor communication generates mistrust, lack of access to mosquito habitats, higher mosquito populations, and the District ultimately having to use less "environmentally friendly" mosquito management strategies in order to meet its mission of protecting public health. Similarly, when members of the public, landowners, and resource agencies narrowly interpret environmental and health laws poor communication and the ability to effectively manage sensitive habitats, species of concern, and mosquito issues can be hindered. The need for a change in perceptions and beliefs, continuous effective coordination and teamwork, broader interpretation and less dogmatic implementation of regulations, and better integration of the many laws with seemingly different missions and purposes, is essential if all parties concerned are to achieve their objectives with minimal detrimental impacts to each other (Cookingham, 1971; Cottam, 1938; Dill, 1990; Eldridge, 1993; Lopp, 1965; Roberts, 1993). The District acknowledges this issue is not new and in fact continues to pose ever increasing challenges, especially with changes in the different agencies' staffing and their subsequent interpretation and implementation of the various regulations. Should the issues of ineffective communication, fragmented cooperation, and narrow dogmatic interpretation of laws persist, the District would be relegated to serving in an advisory role concerning mosquito management in and around sensitive habitats or in areas with species of concern.

The District takes its authority to proactively manage mosquito populations and protect public health, while also meeting its goal of preserving natural resources, very seriously. The District was created and performs its duties pursuant to the Mosquito and Vector Control District Law (Health and Safety Code, §2000 et seq.). In enacting that law the California Legislature recognized the importance to public health and the economy of active management of pests. The Legislature thus found and declared:

Health and Safety Code, § 2001

(1) California's climate and topography support a wide diversity of biological organisms.

(2) Most of these organisms are beneficial, but some are vectors of human disease pathogens or directly cause other human diseases such as hypersensitivity, envenomization, and secondary infections.

(3) Some of these diseases, such as mosquito-borne viral encephalitis, can be fatal, especially in children and older individuals.

(4) California's connections to the wider national and international economies increase the transport of vectors and pathogens

The Legislature granted the District broad powers to address the threat to public health and the economy posed by vectors and specified its duties as follows:

Health and Safety Code, § 2040

Within the district's boundaries or in territory that is located outside the district from which vectors and vector-borne diseases may enter the district, a district may do all of the following:

(a) Conduct surveillance programs and other appropriate studies of vectors and vectorborne diseases.

(b) Take any and all necessary or proper actions to prevent the occurrence of vectors and vector-borne diseases.

(c) Take any and all necessary and proper actions to abate or control vectors and vectorborne diseases.

(d) Take any and all actions necessary for or incidental to the powers granted by this chapter.

Notwithstanding this grant of power, the law does not mandate action by the District and provides that landowners and land managers ultimately are responsible for the abatement of vector populations that breed on their properties and affect public health (Health and Safet Code, § 2060.). Therefore, the District could, if left with no other options concerning mosquito management in sensitive habitats and areas with species of concern or in those parts of communities opposed to mosquito abatement activities, serve merely in an advisory role providing information on the biology of different mosquito species and the different strategies for their management. Unfortunately, passive mosquito management is generally ineffective and results in a very vocal and at times angry citizenry which can create numerous additional challenges not only for the District, but also for landowners/managers, politicians, and resource agencies (Barnard 2013, Colangelo, 2011a,b; DeBenedetto, 2012; DelVecchio and Reed, 1993; Dremann, 2012; Hallissey, 2003; Kuczka, 2002; O'Neill, 1995; Serant, 1998; Terry 2013a,b; Yee, 2013). Furthermore, reactive and/or limited management of mosquitoes by individuals and agencies that are unfamiliar with mosquito habitat, biology, and management strategies is not only inefficient, it can also have unwanted environmental consequences, the least of which is the incorrect and/or excessive use of pesticides and higher incidences of disease and discomfort. Therefore, good communication, coordination, shared compromise, and a blending of the different agency missions is essential to effectively manage mosquito populations, public health, and natural resources.

Tedesco et al. (2010) discusses the importance of communication, coordination, and proactive mosquito management. Their study examined how the local politics of mosquito control affected the spread of West Nile virus in certain Chicago suburbs/villages during the 2002 outbreak. They found the political, economic and philosophical differences between four mosquito abatement districts, coupled with differences in funding, tax base and public and agency attitudes about the environmental impacts of mosquito spraying, played an important role in the preparedness and response capabilities of the four agencies and ultimately in the differences in the number of West Nile virus cases observed. Two agencies enacted aggressive and timely mosquito control and education policies and practices and had significantly fewer human cases of West Nile virus than the two districts that took a more limited and reactive approach. They concluded that the differences in mosquito control practices among the four mosquito abatement districts revealed the interrelated effects of location, scale and politics on West Nile virus management and control. Communication, coordination and timely action with the public, health departments, leaders of suburbs/villages, and with neighboring mosquito abatement districts was essential to successfully reduce the risk of West Nile virus infection.

The inter-connectedness and complexity of managing mosquito populations, protecting public health, and conserving natural resources within the District's Program Area cannot be understated. There are numerous special interest groups as well as members of the public with a wide array of concerns and viewpoints about managing environmental resources, mosquito populations, and public health. There are also numerous local, state and federal regulations, some of which seem to conflict with the proactive management of mosquito populations and the protection of public health. The Legislative findings regarding the importance of active mosquito management and the delegation of power to the District, and its practical experience with reactive management, favor active management, for the benefit of the environment, economy and public health. This active management must be proactive in its approach in order to be an effective IPM program that also protects public health and conserves natural resources.

3.2 Criteria

The District has a well-defined process for selecting tools to be used in mosquito control. The criteria used for determining the viability and ranking of reasonable tools are listed below:

> **Criterion 1.** The District uses tools known to be effective for managing mosquito species that have developed breeding populations in the state.

> **Criterion 2.** The District does not use experimental or hypothetically effective tools.

> **Criterion 3.** Given equal efficacy and operational constraints, the District will use the least environmentally disruptive tool in its control Program.

3.3 Tool Selection Guidelines

The following guidelines (i.e., additional considerations) are used when applying criteria above to the potential mosquito management tools:

> Are there effective control measures for the target mosquitoes or closely related species?

> Are these tools available for use in California?

> Are these tools likely to be effective if used in the District's Service Area?

> Are there environmental circumstances that will likely limit the effectiveness or operational aspects of the tools in natural, rural, or urban settings?

> Are there operational constraints that will limit the effectiveness of the tools?

3.4 Evaluation Results

Table 3-1, Screening with Criteria, shows the results of the scoring for each of the 19 tools described in Section 2 for the three key criteria listed in Section 3.2. Then Table 3-2, applies the tool selection guidelines to those tools meeting program criteria. Some alternatives were eliminated from the analysis because they were infeasible or did not meet the Program's overall objectives, or would not meet the criteria and guidelines for selection. This section concludes with a discussion of how tools remaining (i.e., those following screening with the criteria and the guidelines) were refined further.

Nine tools not passing the Table 3-1 screening were eliminated from further analysis. They are highlighted in Table 3-1: biological control pathogens (viruses), biological control parasites, biological control plants, mass trapping, attract and kill, inundative releases (parasites), inundative releases (predators), regulatory control, and repellents. They are not feasible tools for inclusion in the District's areawide Program at present with one exception under "attract and kill": Attractive Toxic Sugar Bait (ATSB).

Table 3-1 Screening with Criteria

	Criteria		
	1	2	3
Alternative Tools	Method Known to be Effective?	Not Experimental or Hypothetical?	Least Environmentally Disruptive?
Integrated Pest Management	Y	Y	Y
Mosquito Surveillance	Y	Y	Y
Physical Control	Y	Y	Y
Vegetation Management	Y	Y	Y
Biological Control Pathogens (Viruses)	N	N	N/A
Biological Control Pathogens (Bacteria)	Y	Y	Y
Biological Control Parasites	N	N	N/A
Biological Control Predators	Y	Y	Y
Biological Control Plants	N	N	Y
Synthetic Insecticides	Y	Y	N
Natural Insecticides	Y	Y	N
Insect Growth Regulators	Y	Y	Y

Table 3-1 Screening with Criteria

Alternative Tools	Criteria 1: Method Known to be Effective?	Criteria 2: Not Experimental or Hypothetical?	Criteria 3: Least Environmentally Disruptive?
Mineral Oils/Surfactants	Y	Y	N
Mass Trapping	N	N	N/A
Attract and Kill	N	Y	N
Inundative Releases (Parasites)	N	N	N/A
Inundative Releases (Predators)	N	N	N/A
Regulatory Control	Y & N	Y	Y
Repellents	N	Y	Y

Shaded cell indicates that one or more of the criteria were not met and the tool was eliminated from additional analysis.

Y = Yes
N = No
N/A = Not Applicable

ATSBs for the management of adult mosquitoes show promise but are limited in their availability. The District is aware of one new commercially available product, Terminix® AllClear, which contains an essential oil of garlic. The District still needs to operationally test this material, as well as other potential ATSBs, to determine those circumstances where their use may be effective while also having little or no nontarget species impacts. The other 10 tools that passed the initial screening were further analyzed against the guidelines shown in Table 3-2.

Table 3-2 Tool Selection and Application Guidelines

Alternative Tools	Guidelines: Effective Control Measures?	Tools Available in California?	Tools Effective in District Service Area?	Environmental Circumstances Limiting Effectiveness or Operational Aspects of Tools?	Operational Constraints Limiting Effectiveness of Tools?
1. Integrated Pest Management	Y	Y	Y	N	N
2. Mosquito Surveillance	Y	Y	Y	N	N
3. Physical Control	Y	Y	Y	Y	Y
4. Vegetation Management	Y	Y	Y	Y	Y
5. Biological Control Pathogens (Bacteria)	Y	Y	Y	Y	Y
6. Biological Control (Predators)	Y	Y	Y	Y	N
7. Synthetic Insecticides	Y	Y	Y	N	Y

Table 3-2 Tool Selection and Application Guidelines

Alternative Tools	Guidelines				
	Effective Control Measures?	**Tools Available in California?**	**Tools Effective in District Service Area?**	**Environmental Circumstances Limiting Effectiveness or Operational Aspects of Tools?**	**Operational Constraints Limiting Effectiveness of Tools?**
8. Natural Insecticides	Y	Y	Y	N	Y
9. Insect Growth Regulators	Y	Y	Y	N	Y
10. Mineral Oils/Surfactants	Y	Y	Y	Y	Y

Shaded cell indicates that guideline cannot be accomplished in some situations.

Y = Yes
N = No
N/A = Not Applicable

Although some of the tools in Table 3-2 may have environmental and/or operational constraints that limit their effectiveness in some circumstances (as explained below), they remain as viable options and, therefore, are included as components of the District's IMMP. Even with their constraints in some situations, their use meets the District's objective of effectively performing its work of minimizing human-mosquito contact with the least or no environmental risk.

Physical control is a valuable strategy that in many instances results in long-term control of mosquito populations. Over time this strategy results in a significant reduction in pesticide use as well as a cost savings to the District with respect to labor and materials. Additionally, disturbance to sensitive habitats (e.g., tidal marshes) is less, as the need for repeated treatments for breeding mosquitoes is significantly reduced. The limitations vary as follows: (1) Maintenance of recirculation ditches and water control structures in marshes, especially tidal marshes, not only requires permitting from various federal, state, and local regulatory authorities but also has restrictions with respect to the timing of the work so as not to impact the breeding seasons and/or habitats of sensitive species (e.g., California clapper rail and salt marsh harvest mouse) resulting in a very narrow window within which to perform the work (typically September through January of the following year); (2) Height of dredge spoils from ditch maintenance in marshes, especially tidal marshes, is limited to a maximum of 6 inches to prevent the establishment of opportunistic invasive weeds (e.g., pepperweed) and, therefore, sometimes results in performing ditch maintenance annually or biennially to minimize the volume of dredge spoils generated and creation of potential invasive weed habitat; (3) Heavy equipment access can compact soils and damage plants, especially in tidal marsh areas; and, (4) Exclusionary practices are limited in that they are designed to prevent entry of mosquitoes into residences and buildings and do not work well when people and their pets are outside, nor does exclusion work well when people are not disciplined about maintenance of exclusionary structures (e.g., screens), securing points of entry, or taking timely action to engage in exclusionary practices to minimize potential contact with mosquitoes.

Vegetation management is another important component of the District's IMMP that facilitates access to mosquito habitats, reduces the quality of mosquito habitats, and reduces pesticide use. Again, its use has some limitations. First, the use of hand tools may require state and/or federal permits, especially in riparian corridors and tidal marshes because of the presence of special status species and/or the sensitive nature of the habitat, which may be critical for a species of concern. For example, work cannot be performed during Ridgway's rail breeding season. Nor can work degrade the quality of a wetland (e.g., result in stream bank erosion or allow for the establishment of invasive weeds such as arundo, pepperweed, or nonnative spartina). Second, the use of herbicides has limitations with respect to (1) timing of applications for maximum effectiveness (e.g., density, height, species of weed being treated, and time of year), (2) weather restrictions (e.g., wind less than 5 mph to minimize potential drift to unintended targets), and (3) proximity of sensitive habitats and agricultural crops (e.g., vineyards). The use of herbicides may also require an NPDES permit and costly monitoring when used in habitats that are waters of the U.S. or the state. Third, burning is limited to those days during late fall and winter when burning can occur and it is not a designated “spare the air day”. Burning also requires a permit and close coordination with local fire departments and the Bay Area Air Quality Management District (BAAQMD). Lastly, burning can only occur in situations with little risk of the fire escaping from the burn site, the site is not a sensitive habitat or in close proximity to a sensitive habitat, and no special-status species are in or adjacent to the burn site.

Biological control using the bacterial pathogen Bs is an effective tool for controlling immature mosquitoes. Limitations exist though in that some mosquitoes (e.g., those species belonging to the genera *Anopheles*, *Culiseta,* and *Culex*) are more susceptible than others such as *Aedes* spp, especially those found in salt marsh habitats (de Barjac and Sutherland 1990; Baumann et al. 1991; Brown et al. 1991, 2004; Davidson 1989; Lacey et al. 1986, 1988; Lacey and Singer 1982; Mittal 2003; Mulla et al. 1984b, 1985; Ramoska et al. 1978). Other limitations that impede effectiveness include potential for resistance due to over use or reliance on this material as a primary control strategy (Adak et al. 1995; Mittal 2003; Nielsen-LeRoux et al. 1995; Rao et al. 1995; Rodcharoen and Mulla 1994, 1996; Silva-Filha et al. 1995), excessively dense vegetative cover that prevents effective application of the liquid and sometimes the granular formulations to water-breeding mosquitoes, low temperatures from late fall to early spring (Mittal 2003; Mulla et al. 1990b), aquatic habitats with a pH of 10 or greater (Mittal 2003), late fourth stage larvae, which typically do not feed and therefore do not ingest the bacterium, and the need for the presence of dead mosquito larvae for adequate recycling of the Bs for sustained control to occur for up to 4 weeks past the time of the initial application. Also note that a number of other potential biological control agents have been discovered and tested over the years but none of these agents are commercially available at this time.

Biological control using predators is especially limited as very little is commercially available at this time. The predator that is available and that has been used for more than 80 years in California mosquito control efforts is the mosquitofish (*Gambusia affinis)*. Although highly effective, the mosquitofish is a generalist predator and, therefore, does feed on some of the other invertebrate prey present in any given mosquito-breeding habitat. Other factors can limit effectiveness or restrict the use of mosquito-fish. First, water quality. Mosquitofish in general are very hardy and adaptable to a wide range of environments. Still, temperature, dissolved oxygen, amount of organic material in the water, and salinity can all affect reproductive capacity and survival. Second, mosquitofish are less effective in short-term seasonal water habitats than permanent impoundments primarily because they are not able to produce multiple broods and establish a population level that continually regulates mosquito production. Third, density of emergent vegetation can limit fish access to mosquito larvae. Lastly, mosquitofish are a nonnative fish and, therefore, it is important to be careful when placing them in mosquito-breeding habitats or dispensing them to private citizens. California Department of Fish and Wildlife regulations prohibit private citizens from planting nonnative fishes into waters of the state without a permit (see Title 14 California Code of Regulations, Fish and Wildlife Code Sections 1.63, 6400, and 238.5). The District takes care to verify that fish given to citizens are placed only in ornamental water gardens, backyard decorative ponds, large fountains, and horse troughs. Fish are not placed in creeks, streams, marshes, or other types of wetlands or waters of the state.

Synthetic insecticides, the least preferred method for managing mosquito populations, has operational constraints that can limit their effectiveness. First, these materials have the potential to impact nontarget organisms, especially other invertebrates and, therefore, the District is careful when using these materials. For example, the synthetic pyrethroid etofenprox is applied as a fog for adult mosquito control when a temperature inversion and little or no wind is present. Therefore, applications typically occur during the early morning hours up until about 7:30 am or in the late evening hours, depending on temperature and wind speeds. This timing maximizes the presence of the fog to control adult mosquitoes as the length of time the insecticide fog is present is less than about 20 minutes before it dissipates due to settling and/or evaporation of the microdroplets, which are typically smaller than 25 microns in size. It also minimizes unintended drift and reduces potential contact with bees, butterflies, and other nontarget organisms, which typically are not active during the time of application. Second, the proximity of sensitive receptors (e.g., chemically sensitive individuals) requires the District to take every precaution to minimize the risk of exposure. Third, since fogging applications typically occur during times of little or no light, the District is careful to minimize the risk of unintentionally startling livestock, which could injure themselves, or of hitting a landowner's pet that might run out in front of or chase the vehicle while on the property. Thus, the District coordinates its work with landowners whose properties are being fogged in an effort to help them protect their pets and livestock. Fourth, the District uses the granular organophosphate temephos as a mosquito larvicide primarily to combat resistance and in locations where the potential for introduction to unintended habitats and exposure to nontargets can be effectively managed.

Natural insecticides used by the District (both botanical and bacterial) have a number of operational limitations that can limit effectiveness. The pyrethrins that are applied as a fog for adult mosquito control are subject to the same operational conditions and limitations as described in the prior paragraph for the synthetic pyrethroid etofenprox and, therefore, will not be discussed any further. The larvicide Bti is affected by a variety of biological and environmental factors (Lacey 1985) and has the following constraints. First, timing is critical for Bti to be effective as late stage fourth instar larvae typically do not feed and, therefore, will not ingest the larvicide (de Barjac and Sutherland 1990). Additionally, the pupae and adults of mosquitoes are unaffected. Second, the density of vegetative cover can create challenges that prevent effective application of the liquid and sometimes the granular formulations from reaching water-breeding mosquitoes (de Barjac and Sutherland 1990). Third, low temperatures reduce efficacy (Christiansen et al. 2004; de Barjac and Sutherland 1990; Mittal 2003; Wraight et al. 1981). Fourth, high larval populations limit Bti effectiveness (Aly et al. 1988; de Barjac and Sutherland 1990; Becker and Ludwig 1983; Mulla et al. 1982b). Fifth, high organic content or highly polluted waters negatively impacts the usefulness of Bti (de Barjac and Sutherland 1990; Ramoskaet al. 1982). The larvicides developed from *Saccharopolyspora spinosa* are subject to the following limitations that reduce effectiveness: (1) sites with high organic content as the insecticide is readily absorbed and binds to particulate matter making it less available for larval mosquito contact or ingestion (Hertlein et al. 2010); (2) sites subject to full sunlight as the half-life in water exposed to summer sunlight is 1 to 2 days (Hertlein et al. 2010); (3) sites with high water flow, which results in excessive dilution and sublethal dosing of mosquito larvae; and (4) dense vegetative cover, which limits the ability of liquid and even sometimes granular formulations from reaching the water containing mosquito larvae. Also, concern is associated with exposure of bees to spinosyns and, therefore, the formulations used and timing of applications are adjusted to minimize any potential risk to bee populations.

The **insect growth regulator methoprene** is subject to some of the same limitations as the larvicide Bti. First, timing is critical, especially for those species of mosquitoes (e.g., the winter salt marsh mosquito *Aedes squamiger*) whose larvae may take several weeks or even months to complete their development. Applying methoprene too early results in reduced effectiveness because the larval stages effectively metabolize the IGR, which would have prevented adult emergence. Additionally, methoprene does not affect pupal or adult mosquitoes. Second, density of vegetative cover can limit the ability to effectively apply the material to water containing mosquito larvae, especially liquid formulations.

Mineral oil/surfactant larvicides and pupacides are a useful tool that is also a least preferred method for managing mosquito populations. Some issues limit their use and effectiveness. First, similar to the larvicides Bti, Bs, and methoprene, the density of vegetative cover can significantly reduce the effectiveness of these materials as it prevents them from reaching the water where late stage larvae and pupal mosquitoes exist. Second, dense emergent vegetation and/or excessive flotage prevents the spread of surfactants again reducing their efficacy. Third, excessive or continuous wind post-application can render mineral oil/surfactant larvicides and pupacides ineffective as these materials tend to be blown to one side of the treated area and will remain there until the wind activity ceases. Fourth, mineral oil/surfactant mosquitocides can also impact other air-breathing invertebrates, some of whom may also be predators of mosquito larvae. The District makes every effort to encourage the presence of natural predators, as they can help reduce the need to use other pesticides to manage mosquito populations to a healthful level.

3.4.1 Alternatives Considered and Withdrawn from Evaluation

The District determined that of the 19 potential tools, the following 7 were not immediately available or viable for use in its IMMP: biological control pathogens (viruses), biological control (parasites), biological control plants; inundative releases (predators), inundative releases (parasites), mass trapping, and repellants. The first four tools have been withdrawn from further evaluation as they are not commercially available for the District to use. Therefore, they are not viable tools. Mass trapping is not viable for the following reasons: (1) the staff time, and equipment required are exceptionally cost prohibitive, and (2) depletion trapping of mosquitoes, has been shown to be highly ineffective. The use of repellants also has limitations (see Section 2.17 above).Two other tools, attract and kill and regulatory control, have little or no significant effectiveness in managing large mosquito populations. Further analysis of one attract and kill formulation, the new AllClear ATSB, is needed prior to using it for mosquito control. Further analysis of the other forms of attract and kill and regulatory control was deemed unnecessary.

In summary, the District determined that of the 19 potential tools from Table 3-1, the following 9 methods were not immediately available or viable for use in its IMMP: biological control pathogens (viruses), biological control (parasites), mass trapping, attract and kill, inundative releases (both parasites and predators), regulatory control, and repellents.

> *Biological Control Pathogens (viruses)* is deemed infeasible for mosquito control at present. This method is not commercially available in California, and there are currently many efficacy related issues.

> *Biological Control (parasites)* is deemed infeasible as this method is not commercially available in California. Research on the use of parasites for mosquito control has also shown several limitations related to efficacy. Although the use of parasites as a means for managing mosquito populations shows promise, much work concerning their biology, cultivation, mass production, transport, and release remains to be done.

> *Biological Control Plants,* or carnivorous plants, whether terrestrial or aquatic, use a wide range of invertebrate prey and are not specific predators of mosquitoes. What little data exist indicates that carnivorous plants, especially terrestrial species, are inefficient for the control of mosquitoes.

> *Mass Trapping* is not considered by the District to be a practical, effective, reliable method of controlling mosquito populations. Operational difficulties exist in placing out and retrieving large numbers of traps for most mosquitoes, the least of which are the volume of traps required, numbers of staff, amount of staff time, access, and travel necessary for this tool to be effective. Mass trapping of mosquitoes has proven to be both costly and in most instances ineffective.

> *Attract and Kill* is not considered by the District to be a practical, effective, reliable, method of controlling mosquito populations. The technology for mosquitoes is limited, and effectiveness is either not obtained or is inconsistent. Nontarget insects can be impacted. The District is aware of one commercially available ATSB product, Terminix® AllClear. The District still needs to operationally test this material, as well as other potential ATSBs, to determine those circumstances where their use may be effective while also having little or no nontarget species impacts.

> *Inundative Releases of parasites* is not considered by the District to be a practical or currently feasible method of controlling mosquito populations. They are not commercially available and remain experimental at this time.

> *Inundative Releases of predators,* either sterilized or genetically altered, is not considered by the District to be a practical or a currently feasible method of controlling mosquito populations. Introduced species may out compete natural predators and have nontarget species impacts. The use of any genetically altered organisms, even mosquitoes, may not be acceptable to the public.

> *Regulatory Control* is not considered feasible because adoption of regulations is lengthy, time intensive, expensive and uncertain as to the regulatory outcome. This approach is not focused sufficiently on control of existing populations. Moreover, regulatory controls are dependent upon state and federal agencies to initiate and implement, and thus this approach cannot assure that any project objectives would be achieved. Additionally, regulatory actions have the potential to create as well as eliminate additional mosquito habitats.

> *Repellents*, although effective for small-scale use by humans and animals, are not part of the overall Program control strategy because they merely displace the problem and do not reduce the mosquito population in an area. Repellents also require proper application, timely use, and discipline concerning their use to achieve optimal effectiveness. Unfortunately, the use of repellents does not guarantee the elimination of human-mosquito interactions and potential mosquito-borne disease transmission.

3.4.2 Selected Tools and Delivery Techniques

Integrated pest management is an overall approach to the District's Program use of chemical and nonchemical control methods. The following nine tools or "alternatives" were determined to be effective for mosquito control activity: surveillance, physical control, vegetation management (physical, -herbicides, and burning), biological control pathogens (bacteria), biological control predators (specifically fish), synthetic insecticides, natural insecticides, IGRs, and mineral oils and surfactants. For these nine tools, further identification and/or evaluation of the options including how to deliver the material is provided below. Each selected tool will be evaluated further for environmental impacts in the PEIR. Additional discussion of the selected tools (as components of the Proposed Program) is provided in Section 3.5.

3.4.2.1 Evaluation of Material Delivery Options

3.4.2.1.1 Mosquito Larviciding Techniques and Equipment

Due to the wide range of mosquito-breeding sites within the District Service Area, and the pesticide formulations used, the District uses a variety of techniques and equipment to apply larvicides, including handheld sprayers and spreaders, truck- or ATV-mounted spray rigs, and fixed and rotary winged aircraft.

Ground Applications

The District uses conventional pickup trucks and ARGO and Polaris (ATVs) as larvicide vehicles. A chemical container tank, high-pressure, low-volume electric or gas pump, and spray nozzle are mounted in the back of the vehicle bed, with a switch and extension hose allowing the driver to operate the equipment and apply the larvicide. The ATVs may also have booms in lieu of hoses and spray rigs, which allows for applications while steering the vehicle. ATVs are ideal for treating areas such as agricultural fields, pastures, and other offroad sites. Thorough training in ATV safety and handling, as well as minimizing damage to wetlands habitats and sensitive species, is provided to employees before operating these machines.

Additional equipment used in ground applications includes handheld sprayers and backpack blowers. Handheld sprayers (hand cans) are standard 2- or 3-gallon garden style pump-up sprayers used to treat small isolated areas. Backpack sprayers are gas-powered blowers with a chemical tank and calibrated proportioning slot. Generally, a pellet or small granular material is applied with a backpack sprayer or "belly grinder" machine designated to distribute pellets or granules.

Using ground application equipment, both when on foot and when conveyed by vehicles, has several advantages. Ground larviciding allows applications while in close proximity to the actual treatment area and, consequently, treatments to only those microhabitats where larvae are actually present. It also reduces both the unnecessary pesticide load on the environment and the financial cost of the amount of material used and its application. Both the initial and the maintenance costs of ground equipment are generally less than for aerial equipment. Ground larviciding applications are less affected by weather conditions than are aerial applications.

However, ground larviciding is impractical for large or densely wooded areas, or when large numbers of different areas are simultaneously producing mosquitoes. Damage may occur from the use of a ground vehicle in some areas. Unintentional ruts and vegetation damage may occur, although both of these conditions are reversible and generally short lived. Technicians are trained to recognize sensitive areas and to use care and good judgment to avoid significant impacts.

Aerial Applications

When large areas are simultaneously producing mosquito larvae at densities exceeding District treatment thresholds, then the District may use helicopters or other aircraft to apply mosquito larvicides. The District contracts with independent flying services to perform aerial applications, with guidance to the target site provided by District staff. Aerial applications of larvicides is a relatively infrequent activity for the District, typically occurring only a few times each year/once every few years, with each application covering a few hundred acres on average. However, larval production can vary substantially and the District is capable of undertaking more frequent or extensive operations.

Using fixed or rotary wing (helicopter) aerial larvicide application equipment has four advantages compared to ground application. First, it can be more economical for large areas with extensive mosquito production. Second, by covering large areas quickly, it can free District staff to conduct other needed surveillance or control. Third it can be more practical for remote or inaccessible areas, such as islands or large marshes, than ground larviciding. Fourth, no risk exists of temporary damage to the habitat being treated (e.g., tracks, crushed plants) as sometimes occurs with the use of ground equipment. However, risk of drift is greater with aerial applications, especially with liquid or ULV aerial larviciding and, consequently, more risk of nontarget exposure. In addition, accuracy in hitting the target area temporarily requires additional manpower for flagging or expensive electronic guidance systems, which can increase costs. Finally, in addition to the timing constraints inherent in most larvicide use, the potential application window can be very narrow for aerial activities due to weather conditions.

3.4.2.1.2 Mosquito Adulticiding Techniques and Equipment

The District applies adulticides, when needed, primarily from truck-mounted ULV aerosol equipment and, occasionally, from handheld, or ATV-mounted ULV equipment. Adulticide application from the air is possible, but would be used by the District only in emergency conditions. Therefore, aerial aerosol applications are not evaluated.

ULV aerosol machine ("cold foggers") use a forced air blower to generate a fine mist of technical (pure) or highly concentrated insecticide. ULV machines come in a wide variety of sizes, and 8- to 12-horsepower blowers are most common. Unlike earlier "thermal foggers," ULV sprayers use no oil diluent, and their name is derived from the very low volumes of total material sprayed per acre treated. In most mosquito control ground adulticiding operations, application rates rarely exceed 1 ounce per acre, with the particle sizes ranging from 8 to 15-microns.

The sprayers today use several techniques to meet these requirements. Air blast sprayers, which use either high-volume/low-pressure vortical nozzles or high-pressure air-shear nozzles to break the liquid into very small droplets, are most common. Other forms of atomization equipment include centrifugal energy nozzles (rotary atomizers), which form droplets when the liquid is thrown from the surface of a high speed spinning porous sleeve or disc, ultrasonic equipment, which vibrates and throws the droplets off, and electrostatic systems, which repel the droplets.

The insecticide metering equipment available on these machines ranges from an electric pump on fixed flow machines to computer-controlled, speed-correlated, event-recording, and programmable flow management systems. The fixed flow units are designed for operating with the vehicle traveling at a constant speed. Most of these use 12-volt laboratory type pumps, which are quite accurate.

Ground adulticiding equipment is normally mounted in some type of vehicle, but the District also has smaller units that can be carried by hand or on a person's back for small area treatments. Pickup trucks are the most common conveyance for ULV sprayers, but the District can also use ATVs. With the 8- to 12-horsepower midsize sprayers described above, a vehicle speed of about 10 mph typically generates an acceptable dose rate.

The advantages of ULV adulticiding are twofold. First, a large area can rapidly be covered in a very short amount of time. Second, quick relief can be provided by the rapid knockdown of the mosquitoes present, thereby minimizing human-mosquito interactions and any potential for the transmission of mosquito-borne disease. Conversely, the disadvantages are that adulticiding can impact other nontarget organisms (e.g., flies, bees), especially if the application occurs during daylight hours when these other organisms are active. Additionally, adulticiding is temporary, meaning that only those mosquitoes that are present in the target area at the time of application will be affected. Mosquitoes that fly in from neighboring areas or that emerge post-application will be unaffected. Therefore, the District uses adulticiding when all other tools and strategies have been deemed ineffective to manage high mosquito population levels or minimize disease transmission.

3.4.2.1.3 Management of Other Invertebrates

In addition to mosquito control activities, the District may also apply insecticides to control ground-nesting yellow jackets that limit access to mosquito breeding sites. The District does not control any yellow jackets that are located inside or on a structure. If District criteria indicate that a technician should treat a yellow jacket nest, they will apply an adulticide directly to the insect and/or nest to avoid any drift and harm to other organisms.

Pyrethroid based handheld aerosols can rapidly knock down active adults at the nest opening. Dusts containing the active ingredient pyrethrin, can then be blown directly into the active nest using a handheld bulb duster. The potential environmental impacts of these materials is very small because (1) of the active ingredients contained, and (2) the mode of application at the opening or deep into underground nests further limits the potential for environmental exposure from these materials.

3.5 Selected Program Alternatives

The District has selected a systems approach over several years using multiple tools and depending upon conditions at specific locations. The District adopts an overall IPM approach to use procedures that will minimize potential environmental impacts. The District’s Program employs IPM principles by first determining the species, distribution, and abundance of mosquitoes through evaluation of public service requests and field surveys of immature and adult mosquito populations and, then, if the populations exceed predetermined criteria, using the most efficient, effective, and environmentally sensitive means of control. For all mosquito species, public education is an important control strategy. In some situations, water management or other physical control activities can be instituted to reduce breeding sites. The District also uses biological control such as the planting of mosquitofish in some settings: ornamental fish ponds, water troughs, water gardens, fountains, and neglected swimming pools. When these approaches are not effective, or are otherwise deemed inappropriate, then pesticides are used to treat specific mosquito-producing or mosquito-harboring areas.

Three core tenets are essential to the success of a sound IMM program.

> *First*, a proactive approach is necessary to minimize impacts and maximize successful mosquito management. Elements such as thorough surveillance and a strong public education program make all the difference in reducing potential human-mosquito interactions.

> *Second*, long-term environmentally based solutions (e.g., water management, reduction of harborage, exclusion, and enhancement of predators and parasites) are optimal as they reduce the potential pesticide load in the environment as well as other potential long- and short-term impacts.

> *Lastly*, using the full array of options and tools (public education, surveillance, physical control, biological control, and when necessary chemical control) in an informed and coordinated approach supports the overall goal of an environmentally sensitive mosquito management program.

The District's Program consists of the following alternatives, which are general types of coordinated and component activities, as described below. The Proposed Program is a combination of these alternatives with the potential to use all of these alternatives in their entirety along with public education as described below.

3.5.1 Surveillance

Mosquito surveillance, which is an integral part of the District's responsibility to protect public health and welfare, involves monitoring mosquito populations and habitat, their disease pathogens, and human-mosquito interactions. Mosquito surveillance provides the District with valuable information on what mosquito species are present or likely to occur, when they occur, where they occur, their abundance, and if they are carrying disease or otherwise affecting humans. Mosquito surveillance is critical to an IMM program because the information it provides is evaluated against treatment criteria to decide when and where to institute mosquito control measures. Information gained is used to help form action plans that also assist in reducing the risk of mosquito-borne disease transmission and the occurrence of discomfort and injury to humans, pets and livestock. Equally important is the use of mosquito surveillance in evaluating the efficacy, cost effectiveness, and environmental impacts of specific mosquito control actions.

3.5.1.1 Mosquito Surveillance

Mosquitoes in nature are distributed within their environment in a pattern that maximizes their survival to guarantee reproductive success. Immature stages develop in water and later mature to winged adults capable of both long- and short-range dispersal. This duality of their life history presents mosquito control agencies with unique circumstances that require separate surveillance strategies for the aquatic versus terrestrial life stages.

Surveillance involves monitoring the abundance of mosquito populations, their habitat, mosquito-borne disease pathogens, and the interactions between mosquitoes and people over time and space. The District routinely uses a variety of traps for surveillance of adult mosquitoes, regular field investigation of known mosquito sources for direct sampling of immature stages, public service requests for larval and adult mosquitoes, and low-ground-pressure ATVs and watercraft to access these sites. The District conducts surveillance by way of a variety of activities that include:

> **Field counting/sampling and use of trapping**, along with the laboratory analysis of mosquitoes, their hosts, and pathogens to evaluate population densities and potential disease threats such as West Nile virus, western equine encephalomyelitis, and Saint Louis encephalitis. Sampling of presence and abundance of mosquito populations tends to occur in areas where the citizenry would have a likelihood of exposure to them or in habitats occurring within the mosquito species' flight range to populated areas. Field counts take place both at immature and adult stages of mosquito development or life cycle. Four kinds of traps, host-seeking traps, light traps, gravid traps, and oviposition traps are used as described below:

- Host-seeking traps use propane or dry ice (carbon dioxide), octenol, human scent, or combinations thereof to attract female mosquitoes seeking a host on which to blood feed. The trap's components include a battery or power source, a low ampere motor/fan combination, and a collection container for holding captured adults. Depending on the trap, it may also include a dry ice container, an LED light source, or propane tank. Examples of this trap type are Encephalitis Vector Survey (EVS) traps, BG Sentinel traps, and the Mosquito Magnet.
- *Light traps* (commonly called New Jersey light traps) use a source of photo-attraction such as an incandescent lamp (25 watt) or fluorescent lamp (7 watt) where mosquitoes are pulled in by the suction provided by an electric (110-volt AC) appliance motor/fan combination. Mosquitoes picked up by the suction are directed downward (via screened cone) inside the trap body to a glass or plastic collection jar containing a 1-inch strip of Vapona, Hot Shot®, or No-Pest® strip (Dichlorvos). The collection jar is enclosed within an expanded metal cage with a hinged door that is padlocked.
- Gravid traps are used to collect adult mosquitoes that have fed on hosts and are seeking a place to deposit eggs. As an example, they may use 5-day-old hay-infused water contained in a small plastic dish pan that has a 6-volt battery-operated fan directly above to draw the gravid female mosquitoes into the small collection net. Another example is the Autocidal Gravid Ovitrap (AGO). This device is a black 5 gallon bucket and lid with a black bottomless 1 gallon bucket inserted in the center. The 5 gallon bucket can hold upto 2.5 gallons of an attractant, such as water and decaying vegetation. The top of the one gallon container is covered with a ¾" mesh netting to allow mosquitoes to enter and exclude large debris. The bottom of the one gallon container is fitted with window screen on the bottom to keep adult mosquitoes from accessing the water in the 5 gallon bucket below. The sides of the 1 gallon bucket are coated with a nontoxic adhesive to capture the adult mosquitoes for identification.
- Oviposition traps are a passive surveillance tool for detecting the presence of container-inhabiting mosquitoes, and for providing a relative measure of temporal changes in adult abundance. These are usually small cups, partially filled with water, with a strip of filter paper just above the water level. Adult females lay their eggs on the surface of the filter paper.

Mosquito immatures include eggs, four larval stages, and a transitional pupal stage. Mosquito control agencies routinely target the larval and pupal stages to preclude an emergence of adults. Operational evaluation of the presence and abundance of immature mosquitoes is limited to the larval and pupal stages, although the District may sample eggs for research reasons. Sampling and collection of the immature stages (egg, four larval stages, and a transitional pupal stage) involves the use of a 1-pint dipper (a standardized small plastic pot or cup-like container on the end of a 36-inch handle), which scoops up a small amount of water from the mosquito-breeding site. Operationally, the abundance of immatures in any identifiable "breeding" source is measured through direct sampling, which provides relative local abundance as the number of immatures per unit volume or area of the source. This method requires access by field personnel to within about 3 feet of larval sites at least every week in warm weather. The spatial patchiness of larvae requires access to multiple locations within each source, rather than to single "bell-weather" stations.

> **"Arbovirus"[2] surveillance to determine the likelihood and occurrence of mosquito-borne illness** is accomplished by three methods commonly used in California: (1) capturing and testing female vector mosquitoes for the presence of mosquito-borne viruses as explained above (2) periodic testing for the presence of encephalitis virus-specific antibodies in the blood serum of sentinel chickens and (3) testing for the presence of encephalitis virus-specific antibodies in the blood serum of domestic or wild birds. The first method involves the use of host-seeking traps to capture female vector mosquitoes. Captured

[2] Arthropod-borne viruses. The primary reservoir for the pathogens that cause these diseases is wild birds, and humans only become exposed as a consequence of an accidental exposure to the bite of infective mosquito s.

females are sorted into groups of up to 50 (called pools) and tested via one of two methods. Testing may be carried out in the District's laboratory using a method such as real-time polymerase chain reaction (RTPCR) or the mosquito pool may be submitted to UC Davis to test for the presence of mosquito-borne viruses. The District uses method (2) above through the placement of caged chickens as “sentinel birds.” Since the viruses of major concern (West Nile virus, western equine encephalomyelitis, and Saint Louis encephalitis) are diseases actively transmitted by mosquitoes to both birds and to humans through bites, caged chickens’ routine blood samples will reveal whether one or more of the virus-specific antibodies are present. The chickens are placed generally 7 to a caged area (at least 6 by 12 feet or larger), are humanely treated, and are provided ample shelter with nest boxes, water, and feed. Chickens are used as the early detection system for virus transmission, as they are unaffected by the presence of these viruses in their systems. At the end of the mosquito season, the chickens are adopted out. Lastly, the District participates in the state's dead bird pickup program as part of its West Nile virus surveillance program. Dead birds deemed suitable for testing are collected or brought to the District and are either tested in-house or sent to a lab for testing.

> **Field inspection of known or suspected habitats** where mosquitoes live and breed is routinely conducted. Sites where water can collect, be stored, or remain standing for more than a few days are potential habitats for mosquito breeding that require continuous inspection and surveillance. Water runoff into catch basins and stormwater detention systems from land uses including, but not limited to, residential communities, parks and recreation areas, and industrial sites, as well as ornamental ponds, unmaintained swimming pools, seeps/seepages, seasonal wetlands, tidal and diked marshes, freshwater marshes, wastewater ponds, sewer plants, winery waste/agricultural ponds, managed waterfowl ponds, canals, creeks, streams, tree holes, tires, man-made containers, flooded basements/crawl spaces, and other standing waters are likely sources.

> **Maintenance of paths and clearings** to facilitate sampling and to provide access to mosquito habitat is an important part of the District's surveillance and control program. It is District policy that staff use preexisting roads, trails, walkways, and open areas whenever possible to conduct routine and essential surveillance activities to minimize impact on the environment. Surveillance is conducted on foot or by using ATVs and low-pressure ground vehicles, with offroad access minimized and used only when roads and trails are not available.

> **Other methods of data collection.** The District’s mosquito surveillance activities are conducted in compliance with accepted federal and state guidelines, in particular the *California Mosquito-borne Virus Surveillance and Response Plan* (CDPH and MVCAC 2012a) and *Best Management Practices for Mosquito Control in California* (CDPH and MVCAC 2012b).These guidelines recognize that local conditions will necessarily vary and, thus, call for flexibility in selection and specific application of control methods.

3.5.2 Physical Control

Managing mosquito habitat to reduce mosquito production or migration, either directly or through public education is often the most cost-effective and environmentally benign element of an IMM program. This approach to the control of mosquitoes and other pests is often called “physical control” to distinguish it from those mosquito management activities that directly rely on application of chemical pesticides (chemical control) or the introduction or relocation of living agents (biological control). Other terms that have been used for mosquito habitat management include “source reduction,” which emphasizes the significance of reducing the habitat value of an area for mosquitoes, or “permanent control,” to contrast with the temporary effectiveness of pesticide applications.[3] Mosquito habitat management is important because its use can reduce or virtually eliminate the need for pesticide use in and adjacent to the affected

[3] This terminology can be misleading if periodic maintenance is needed for physical control devices or structure.

habitat and, in some situations, can virtually eliminate mosquito production from specific areas for long periods of time, reducing the potential disturbances associated with frequent biological or chemical control activities. The intent is to reduce the abundance of mosquitoes produced or sheltered by an area while protecting or enhancing the habitat values of the area for desirable species. In many cases, physical control activities involve restoration and enhancement of natural ecological functioning, including production and dispersal of special status species and/or predators of mosquitoes.

3.5.2.1 Mosquitoes

Physical control for mosquitoes consists of the management of mosquito-producing habitat (including freshwater and tidal marshes and lakes, saltwater marshes, temporary standing water for 4 days or more, and wastewater treatment facilities) especially through water control and maintenance or improvement of channels, tide gates, levees, and other water control facilities. Physical control is usually the most effective mosquito control technique because it provides a long-term solution by reducing or eliminating mosquito developmental sites and ultimately reduces the need for chemical applications. Physical control practices may be categorized into three groups: maintenance, new construction, and cultural practices.

Maintenance activities are conducted within tidal, managed tidal and nontidal marshes, seasonal wetlands, diked, historic baylands, and in some creeks adjacent to these wetlands. The following activities are classified as maintenance:

1. Removal of sediments from existing water circulation ditches
2. Repair of existing water control structures
3. Removal of debris, weeds, and emergent vegetation in natural channels
4. Trimming of brush for access to streams or wetland areas
5. Filling of existing, nonfunctional water circulation ditches to achieve required water circulation dynamics and restore ditched wetlands

New construction typically involves the creation of new ditches to enhance tidal flow preventing stagnant water.

Cultural practices include vegetation and water management, placing culverts or other engineering works, and making other physical changes to the land. These practices reduce mosquito production directly by improving water circulation and indirectly by improving habitat values for predators of larval mosquitoes (fish and invertebrates), or by otherwise reducing a site's habitat value to mosquito larvae.

The District performs these physical control activities in accordance with all appropriate environmental regulations (e.g., wetland fill and dredge permits, endangered species review, water quality review, streambed alteration permits, and in a manner that generally maintains or improves habitat values for desirable species. Major physical control activities or projects (beyond the scope of the District's 5-year regional wetlands permits with the US Army Corps of Engineers San Francisco Bay Conservation and Development Commission, and San Francisco Regional Water Quality Control Board are addressed under the PEIR where known and identified. Minor physical control activities (covered by the regional wetlands permits) are also addressed in the PEIR. They vary substantially from year to year, but typically consist of up to 35,000 linear feet of ditch maintenance. Under the regional permits, the District's work plans are reviewed annually by trustee and other responsible agencies prior to initiation of the planned work. Completed work is inspected by US Army Corps of Engineers, US Fish and Wildlife Service, California Department of Fish and Wildlife (formerly Fish and Game), and other responsible agencies.

The District may request/require landowners and stewards to maintain and clear debris from drainage channels and waterways; excavate built-up spoil material; remove water from tires and other urban containers; cut, trim, mow, and harvest aquatic and riparian plants (but not including any mature trees, threatened or endangered plant species, or sensitive habitat areas); and perform minor trenching and ditching. Requests made of landowners and stewards, exclusive of residential or business situations that involve small manmade containers (e.g., buckets, barrels, wheelbarrows, pots, fountains), include clear recommendations that they consult with the appropriate resource agencies and expert biologists (e.g., USFWS, NMFS, CDFW, BCDC. USACE, State Water Board) as well as acquire any permits prior to commencement of any work. The District makes every effort to assist landowners and managers in protecting sensitive habitats and species while also managing problematic mosquito populations.

3.5.3 Vegetation Management

3.5.3.1 Physical Control

The species composition and density of vegetation are basic elements of the habitat value of any area for mosquitoes, predators of mosquitoes, and for protected flora and fauna. District staff periodically undertake vegetation management activities, or encourage and teach others how to do so on their property, as a tool to reduce the habitat value of sites for mosquitoes or to aid production or dispersal of mosquito predators, as well as to allow access by District staff to mosquito habitat for surveillance and other control activities. Direct vegetation management by District staff generally consists of activities to reduce the mosquito habitat value of sites by improving water circulation or access by fish and other predators, or to allow access by District staff to standing water for inspections and treatment.

For vegetation management, the District uses hand tools and may potentially use other mechanical means (i.e., heavy equipment) for vegetation removal or thinning and could potentially apply herbicides (chemical pesticides with specific toxicity to plants) to improve surveillance or reduce mosquito habitats. Vegetation removal or thinning primarily occurs in aquatic habitats to assist with the control of mosquitoes and in terrestrial habitats to access mosquito breeding sources. To reduce the potential for mosquito breeding associated with water retention and infiltration structures, District staff may systematically clear weeds and other obstructing vegetation in wetlands and retention basins (or request the structures' owners to perform this task). In particular, thinning and removal of emergent vegetation overgrowth would be done to provide a maximum surface coverage of 30 percent or less. In sensitive habitats and/or where sensitive species concerns exist, vegetation removal and maintenance actions would be coordinated with the appropriate resource agencies, have permits, and be restricted to those months or times of the year that minimize disturbance/impacts. Vegetation management may also be performed to assist other agencies and landowners with the management of invasive/nonnative weeds (e.g., spartina, pepperweed, arundo, tamarix, and ailanthus). These actions are typically performed under the direction of the concerned agency, which also maintains any required permits. Any recommendations or requests made to landowners and managers (exclusive of residential yards that do not have or are not part of or adjacent to sensitive areas) also includes the requirement to contact resource agencies and professional biologists concerning the presence of special status species and sensitive habitats as well as the need for acquiring any permits prior to commencement of work.

Tools ranging from shovels and pruners to chain saws and "weed-whackers" up to heavy equipment can all be used at times to clear plant matter that either prevent access to mosquito-breeding sites or that prevent good water management practices that would minimize mosquito populations. Generally, however, District "brushing" activities rely almost entirely on hand tools. Trimmed vegetation is either removed and disposed of properly from the site or placed and/or broadcast in such a way as to minimize visual degradation or impacts to the habitat. Trimming is also kept to a minimum to reduce the possibility of the invasion of exotic species of plants and animals. Surveys for special status plants, coordination with resource agencies and the landowner, and acquisition of necessary permits are completed before any work is undertaken. Follow-up surveys are also conducted to verify that the work undertaken was effective and that the physical manipulation of the vegetation did not result in any unintended overall habitat degradation.

In addition, the use of water management to control vegetation is in some ways an extension of physical control, in that water control structures created as part of a physical control project may be used to directly manipulate hydroperiod (flood frequency, duration, and depth) as a tool for vegetation management. Where potential evapotranspiration rates are high, water management can also become a mechanism for salinity management and, indirectly, vegetation management through another path.

3.5.3.2 Herbicides

Herbicides that may be used by the District to control mosquito populations are listed below. The application of herbicides is the least preferred method for vegetation management and is a last resort when other strategies have been deemed ineffective and/or impractical. All herbicides would be applied in strict conformance with label requirements and any applicable federal and state requirments.

3.5.4 Biological Control

Biological control of mosquitoes involves the intentional use of mosquito pathogens (diseases), parasites, and/or predators to reduce the population size of target mosquito populations. It is one of the principal components of a rational and integrated mosquito control management program. The effectiveness of a mosquito biological control agent lies in its ability to reduce mosquito numbers as quickly as possible. An ideal biological control agent feeds preferentially on the target mosquitoes, exhibits an extremely efficient hunting or parasitizing strategy, and reproduces quickly. Biological control is used as a method of protecting the public from mosquitoes and the diseases they transmit without the use of pesticides and potential problem of pesticide resistance; however, the use of pathogens involves chemical treatment with US Environmental Protection Agency-registered materials. The different types of biological controls are described in the following paragraphs.

3.5.4.1 Mosquito Pathogens (Viruses and Bacteria)

Mosquito pathogens include an assortment of viruses and bacteria. Pathogens are highly host-specific and usually infect mosquito larvae when they are ingested. Upon entering the host, these pathogens multiply rapidly, destroying internal organs and consuming nutrients. The pathogen can be spread to other mosquito larvae in some cases when larval tissue disintegrates and the pathogens are released into the water to be ingested by uninfected larvae. Examples of viruses that can infect mosquitoes are mosquito iridoviruses, densonucleosis viruses, nuclear polyhedrosis viruses, cytoplasmic polyhedrosis viruses, and entomopoxviruses. An example of a bacterium pathogenic to mosquitoes is Bs. The bacterium Bs produces proteins that are toxic to most *Culex* and *Anopheles* mosquito larvae. Bs can reproduce in natural settings for some time following release. Bs is a naturally occurring soil organism that is commercially produced as a mosquito larvicide.

3.5.4.2 Mosquito Predators

Mosquito predators are represented by highly complex organisms, such as insects, fish, birds, and bats that consume larval or adult mosquitoes as prey. Predators are opportunistic in their feeding habits and typically forage on a variety of prey types, which allows them to build and maintain populations at levels sufficient to provide some level of mosquito control, even when mosquitoes are scarce. Examples of mosquito predators include representatives from a wide variety of taxa: coelenterates, *Hydra* spp; platyhelminths, *Dugesia dorotocephala, Mesostoma lingua, and Planaria* spp; insects, *Anisoptera, Zygoptera, Belostomidae, Geridae, Notonectidae, Veliidae, Dytiscidae, and Hydrophilidae*; arachnids, *Pardosa* spp; fish, *Gambusia affinis* and *Gasterosteus aculeatus*; some bats; and some birds belonging to the *anseriformes, apodiformes, charadriiformes, and passeriformes*. Only mosquitofish (*Gambusia affinis*) are commercially available to use at present, while the District supports the presence of the other species as practical.

The District's rearing and stocking of mosquitofish in mosquito habitat is the most commonly used biological control agent for mosquitoes in the world. These fish are ideal control agents for several reasons. They feed primarily at the water's surface, where larvae can be found. Their small size allows them to access vegetated and shallow areas. They can tolerate a significant range in water temperature and water quality. They are also easy to handle, transport, stock, and monitor. Correct use of this fish can provide safe, effective, and persistent suppression of a variety of mosquito species in many types of mosquito sources. As with all effective control agents, the use of mosquitofish requires a good knowledge of operational techniques and ecological implications, careful evaluation of stocking sites, use of appropriate stocking methods, and regular monitoring of stocked fish. Mosquitofish reproduce in natural settings, for at least some time after release. Due to allegations that mosquitofish may potentially impact red-legged frog and tiger salamander populations, District policy is to limit the use of mosquitofish to ornamental fish ponds, water troughs, water gardens, fountains, neglected or unmaintained swimming pools, and other types of artificial man-made habitats. Limiting the introduction of mosquitofish to these sources should prevent their migration into habitats used by threatened, endangered, or rare species. On average, the District releases about 50 pounds of mosquitofish annually.

3.5.5 Chemical Control

Chemical control is a Program tool that consists of the application of limited or nonpersistent selective insecticides (and potentially herbicides as noted earlier in Section 3.5.3.2 above) to directly reduce populations of larval or adult mosquitoes and other invertebrate threats to public health (e.g., yellow jackets) and the use of rodenticides to control rats and mice. If and when inspections reveal that mosquito populations are present at levels that trigger the District's criteria for chemical control – based on mosquito abundance, density, species composition, proximity to human settlements, water temperature, presence of predators, and other factors – District staff will apply pesticides to the site in strict accordance with the pesticide label instructions and any applicable federal and state requirements.

3.5.5.1 Synthetic Insecticides

Synthetic insecticides are pest management products produced in a laboratory and in some cases may also be a synthetic version of naturally occurring pesticides (e.g., pyrethroids that are a synthetic version of naturally occurring pyrethrin). The District sometimes uses the pyrethroids etofenprox and resmethrin, as well as the organophosphate temephos, to help manage mosquito populations. Etofenprox and resmethrin are applied using ULV fogging machines at a maximum rate of less than 0.8 ounce per acre as a mosquito adulticide. Temephos, used only in granular form, is to combat resistance in immature stages of mosquitoes and is applied by hand. The use of synthetic insecticides is the least preferred method due to the potential impacts to nontarget organisms. All of these materials are highly effective but nonselective. Therefore, this tool is used when all other methods are no longer practical or have not been successful in effectively reducing mosquito populations to a healthful level for people, pets, and livestock.

3.5.5.2 Natural Insecticides

Natural insecticides are those materials made directly from plants or other organisms such as bacteria. Some of these materials, such as Bti, are highly host specific, while others such as pyrethrin are not.

Botanical insecticides are derived from plants (e.g., pyrethrins from chrysanthemum flowers). The District sometimes uses pyrethrin to manage adult mosquitoes and potentially yellow jackets. The use of pyrethrin is also a least preferred method for controlling mosquitoes. Pyrethrins break down rapidly (usually within hours) when exposed to sunlight. The District recognizes that pyrethrins are not selective for mosquitoes. Therefore, use near beehives is restricted. Additionally, wind restrictions also apply to minimize unwanted drift when making ULV fogging applications. Pyrethrin for adult mosquito control is applied at a maximum rate of less than 0.8 ounce per acre using a ULV fogging machine. Pyrethrin dust for the treatment of yellow jacket nests would be applied at a maximum rate of 2 ounces per nest and is performed with a handheld bulb duster that blows the pyrethrin directly into the nest.

Insecticides derived from bacteria (e.g., Bti) typically consist of a chemical by-product and/or protein spore produced directly from the organism. The bacterium Bti produces spores containing protein molecules or crystals that are toxic to most immature mosquitoes. The various formulations of Bti used by the District contain no live bacteria but only the spores with protein molecules. Bti efficacy is reduced in highly organic or polluted waters, low temperatures, areas with high larval densities or when dense vegetative cover interferes with application at the mosquito-breeding site*.* Additionally, timing of the application is critical to maximize effectiveness as the adult, pupal, and late 4th instar larval stages of mosquitoes are not susceptible to Bti. Even with the above limitations, Bti is highly effective and, therefore, a preferred method for the management of mosquito populations when predators, biological control, and habitat manipulation strategies are ineffective. This material comes in liquid, granular, powder, and water-soluble packet formulations. The liquid formulations of this insecticide are applied using a hand can, truck-boat, or ATV-mounted sprayer, or by fixed wing or rotary winged aircraft. Granular formulations are applied by hand, or with a granular spreader mounted on a boat or ATV, or via air with fixed wing or rotary winged aircraft. The Bti powders are currently not used by the District. Historically, powdered formulations were mixed with sand and a small amount of mineral oil to act as a binding agent, and then the mix was applied with a handheld granular spreader. The District may in the future use powdered Bti, if and when it becomes available again, and,- therefore,- makes mention of it here to keep it as an available option for larval mosquito control. Water soluble packets are used to help control larval mosquitoes that are present in small containers, ornamental water gardens, stormwater detention systems- and storm drains. Water-soluble packets are only applied by hand.

Another bacterium, *Saccharopolyspora spinosa*, produces compounds known as spinosyns, which are toxic to immature mosquitoes. Like Bti some physical and environmental conditions can limit the effectiveness and use of this material. Unlike Bti, rates near maximum label rates have been shown to affect a few species of nontarget organisms, while lower rates appear to be more specific to immature mosquitoes. Research has demonstrated that mosquito larvae are highly sensitive to spinosyns, although additional research is needed to confirm minimum effective field rates for mosquito control purposes. The District currently uses granular and tablet forms of this material as a part of its IMMP.

3.5.5.3 Insect Growth Regulators

IGRs target immature insect populations. IGRs can be target specific, depending on the formulation used and the concentration that is applied to the target population of insects being managed. Therefore, adhering to label requirements and used in the manner for which they are designed, IGRs affect the juvenile stages of the target organisms while causing little or no effects to the nontargets present (e.g., methoprene and mosquitoes). Unlike many traditional insecticides, IGRs do not affect an insect’s nervous system, nor do they kill adult mosquitoes. Rather, IGRs prevent the ability of the immature stages to complete their final molt from the pupal stage to adult (prevent adult emergence).

Methoprene is a synthetic juvenile hormone that is used by the District to manage mosquito populations. This insecticide is most effective on the pupal and late fourth instar larval stages. It is absorbed on contact and causes an imbalance in the hormone system of the mosquito resulting in its inability to complete metamorphosis to the adult stage. The maximum label rate for application of this insecticide for mosquito control is many magnitudes below the levels that could impact other nontarget organisms, specifically invertebrates, amphibians, and fish, making it a good tool for use in the District's IMMP. Persistence and bioaccumulation in the environment are also insignificant as methoprene readily biodegrades in the presence of ultraviolet light and is also readily metabolized; hence, the timing of applications for this material are essential for optimal effectiveness. The half-life of methoprene is about 2 days in water, 2 days in plants, and 10 days in soil. Formulations used by the District are liquid, pellets, and 30-day briquettes. Both the pellets and briquettes are slow release formulations that allow for concentrations just sufficient to prevent adult emergence of mosquitoes to occur for up to 30 days. Liquid methoprene is applied using hand can, truck-or ATV-mounted sprayers, or by fixed wing or rotary winged aircraft. Granular formulations are applied by hand or with a granular spreader that is mounted on a boat, ATV or carried by aircraft. Briquette formulations are applied by hand to small man-made containers, water gardens, fountains, abandoned swimming pools, stormwater detention systems, and storm drains.

3.5.5.4 Mineral Oils/Surfactants

Mineral oil and ethoxylated alcohol formulations (also known as surfactants) are used to control immature stages of mosquitoes (larvae and pupae). This control is accomplished by changing the surface tension of the water resulting in suffocation. These materials can also affect any adult mosquito that tries to land on the water to rest or lay eggs. Unfortunately, other air-breathing aquatic and semiaquatic insects, including, but not limited to, water beetles, certain flies, water boatman, water striders, and backswimmers, that are exposed to these surfactants can also be affected. Therefore, this tool is used as a last resort to prevent adult emergence when all other immature mosquito control methods are deemed to be ineffective. The current surfactants available are BVA-2 Oil and Coco Bear Oil. Agnique MMF is currently not available for use, although it is possible that its use may change sometime in the future and it is, therefore, included as a part of the District's IMMP. The active ingredient in BVA-2 is mineral oil. Coco Bear Oil is comprised of 10 percent mineral oil with the remaining oil content consisting of food grade coconut and vegetable oils. Agnique MMF is 100 percent ethoxylated alcohol. All of these materials can be applied using a hand can, truck- , boat- or ATV-mounted sprayer, or with a handheld 1-pint spray bottle.

4 No Project Alternative

No Project is defined as what would reasonably be expected to occur in the foreseeable future, based on current plans and consistent with available infrastructure and community services, if the project was not approved and implemented. For the District, the Proposed Program is to continue current activities and introduce similar pesticides to those currently in use if needed in the future. The No Project/No Program condition assumes that the current activities would cease and result in a “do nothing” alternative. It must be evaluated in comparison to the existing condition for CEQA compliance. Key assumptions for the No Project Alternative for inclusion in the District’s PEIR are:

> Current regulatory controls would continue and expand as needed; however, the District would not engage in implementing any of these regulations concerning public health and management of mosquitoes carrying potential diseases. For all practical purposes, the District’s office would close, and public education and other outreach activities would cease along with the control activities.

> Private landowners would manage mosquito problems on private land without any state or federal oversight of pesticides currently registered and available for use. Households would use pesticides commonly available from retail outlets where organophosphates, pyrethrin, and pyrethroids are common ingredients.

> Private landowners would also manage mosquito habitats (clearing, brushing, and draining) with potentially little or no oversight.

4.1 Implications of No Project Alternative

“Doing nothing” as the No Project Alternative has potentially serious implications for public health, economic, and environmental conditions in the District’s Program Area.

4.1.1 Public Health

A wide range of public health issues would occur with the No Project Alternative, First, risk of human cases of mosquito-borne disease and mosquito interaction issues for humans, pets and wildlife would increase. The San Francisco Bay Area has a well-documented history concerning human-mosquito interaction. The earliest written record dates back to the 1772 diaries of Father Juan Crespi who described the "swarms of mosquitoes" in the Warm Springs Area of the City of Fremont and below the hills of Berkeley (Bolton 1927; Gray 1951). Additional records include the 1810 journal entry of mosquitoes attacking a detachment of soldiers near the Albany Hills as well as references indicating that the indigenous peoples of the Bay Area would take action to avoid the large numbers of mosquitoes present during certain times of the year. Note that these interactions took place at a time when the Bay’s wetlands and sensitive habitats were essentially pristine, having limited human habitation and little or no draining, filling or modification, or loss of wildlife including predators of mosquitoes.

Second, the lack of any form of coordinated surveillance reduces the ability of any agency to perform disease risk assessments and, therefore, predict potential outbreaks. Although mosquito-borne disease is not as prevalent as in other areas of the world, mosquito-borne pathogens are still present. West Nile virus, introduced to California in 2003, is present throughout the Bay Area, with positive birds, human cases, and squirrels still detected and reported every year. Malaria continues to be a concern as introduced cases are detected in travelers returning from malaria-infected regions and some recent immigrants every year. The mosquito for this pathogen can be found in many areas of the San Francisco Bay region, and reintroduction of the malaria organism into local mosquito populations is monitored closely. The last known endemic transmission of malaria occurred in the Putah Creek area of Napa and Solano counties in 1939.

Third, lack of coordinated surveillance increases the risk of emerging infectious diseases or mosquitoes going undetected until they have become established. The appearance of West Nile virus in New York City in 1999 is an excellent example. For budgetary and other reasons, New York had significantly reduced their mosquito surveillance and management program many years prior to 1999. By the time the virus had been identified, a number of human cases had already occurred and the virus had become well established. Now the virus is endemic throughout the US and results in numerous cases nationwide. Similarly, the reintroduction of mosquito-borne diseases such as malaria and dengue that had not been present for many years or even decades could also go undetected until their reestablishment or an outbreak of human cases (Brunetti et al. 1954; Gubler and Clark 1995; Maldanado et al. 1990; Radke et al. 2012; Singal et al. 1977).

Fourth, lack of public outreach results in less current information being available about mosquitoes and mosquito-borne disease risk reduction. This lack can lead to increased production of mosquitoes on private property as well as increased cases of mosquito-borne disease in humans, their pets, and livestock. Additionally, the increase in mosquito-human interactions would result in an increased risk of severe reactions to the bites in sensitive and immunocompromised individuals. Research over the last 75 years has documented cases of hypersensitivity and/or severe reactions to mosquito bites in children, immunocompromised individuals, and persons infected with the Epstein-Barr virus or being treated with zidovudine for the AIDS virus. (Brown et al. 1938; Diven et al. 1988; Galindo et al. 1998; McCormack et al. 1995; Peng et al. 2004; Seon et al. 2013; Simmons and Peng 1999; Smith et al. 1993; Weed 1965). Crisp and Johnson (2013) provide a review of mosquito allergy including immunology, diagnosis, and treatment and conclude (1) treatment should focus on avoidance including limiting breeding sites for mosquitoes as well as the use of repellents and protective clothing, (2) local immediate reactions can be managed with the use of prophylactic antihistamines, (3) individuals with severe or anaphylactic reactions to mosquito bites should carry with them Epi-Pens (autoinjectable epinephrine), and (4) more research is needed in a number of areas concerning management and treatment of patients with hypersensitivity to mosquito bites.

The reaction of persons to mosquito bites, clearly brings into question the use of the terms "nuisance" and "pest" that have commonly been used in the past to define the difference between those mosquito organisms that transmit mosquito-borne diseases (i.e., malaria, West Nile virus) and those that do not. The use of these terms is a misnomer and should not be used to characterize the importance of one mosquito species over another. Human-mosquito interactions result in a wide range of mental, emotional, and physical responses, all of which have health implications even in the absence of pathogenic organisms. California Health and Safety Code, Division 104, Part 11, Chapter 1, Section 116108 defines a vector as "any animal capable of transmitting the causative agent of human disease or capable of producing human discomfort or injury including, but not limited to, mosquitoes, flies, other insects, ticks, mites, and rats." This definition inherently recognizes that human discomfort and injury as a result of human-mosquito interactions, is by its own nature, an issue of health just as important as any mosquito-borne agent of human disease.

4.1.2 Economic Conditions

A number of economic issues are associated with the No Project Alternative. First, with increased human-mosquito interactions comes an increase in the number of cases of mosquito-borne disease. The short-term medical and lost workplace, school, and home time associated with illness can cost governments, businesses, families, and individuals upwards of many thousands of dollars (Armien et al. 2008; Barber et al. 2010; Clark et al. 2005; Gubler 2002; Halasa et al. 2012; Meyers 1922; Shepard et al. 2011; Suaya et al. 2009; Tam et al. 2012; Torres 1997; Von Allmen et al. 1979; Vora et al. 2014; Wettstein et al. 2012). For long-term severe cases that result in paralysis, persistent fatigue, muscle weakness, and/or decreases or loss of cognitive function, this cost can mean millions of dollars to families and federal and state governments (Staples et al. 2014; Utz et al. 2003; Villari et al. 1995). Although not as common, no monetary value can be adequately calculated for the loss of life due to mosquito-borne disease. Additionally, the loss of valuable livestock (e.g., horses) and decreased farm productivity can also be significant (Abbitt and Abbitt 1981; ASTHO no date; Byford et al. 1992; Cattell 1916; Gadsen Times 1980; Geiser et al. 2003; Herrick 1903; Hoffman and McDuffie 1963; Howard 1909; Mongoh et al. 2008; Steelman et al. 1973, 1972; Williams et al. 1985).

Second, increased mosquito populations can lead to reduced outdoor recreation activities by the public (Halasa et al. 2014), resulting in increased usage of electricity for air conditioning and indoor entertainment such as television, video games, computers, lighting, etc. These increases could also lead to a reduction in revenues for recreational areas such as parks, campgrounds, marinas, and other areas that depend on usage fees to help with their maintenance and staffing. Outdoor activities are also significant to tourism, which for many areas is an important part of their economy. Large mosquito populations and/or reported cases of mosquito-borne disease can impact tourism and potential revenues (Gaiser 1980; Kirka 1989; Merco Press 2008; The Hindu 2007; Wagner and Magee 1977; Williams 1986).

Third, increased mosquito populations not only lead to increased levels of mosquito-borne disease but can also result in decreased property values (Herms and Gray 1944; Howard 1909). Within San Francisco Bay, historical mosquito populations were at times so severe as to impact real estate sales (Gray 1951). The impact of mosquito control work on property values is also illustrated by Headlee (1945), who summarized the economic effect of mosquito control work in New Jersey. Here property valuations from 1915 to 1930 had increased by $555,345,000.00 over what was expected for those communities that had received mosquito control work. Property values form an essential part of the revenue stream for government services such as schools, police, fire, libraries, parks, and health and welfare programs.

Fourth, the cost of hiring private contractors to provide mosquito control services on a site-specific basis can end up more costly than the costs associated with the current program (with economies of scale). More significant are the costs associated with having to reestablish a program that has been eliminated. These costs include equipment, staffing, staff training, and the initial environmental costs associated with a new program working to restore mosquito levels to the healthful level that existed with the old program prior to its elimination. A loss of institutional memory and understanding of local mosquito populations, their habitats, and the local citizenry cannot be replaced when a program is eliminated. When a program is reestablished, less environmentally friendly measures will be employed during a period of time to bring mosquito populations down to a level where maintenance and control measures that have little or no environmental impact can be effectively employed (e.g., New York and West Nile virus).

4.1.3 Environmental Conditions

The environmental issues associated with the No Program Alternative cannot be understated. First, in the absence of organized mosquito control programs, unlicensed individuals could begin applying over the counter pesticides on their own. Most of these individuals have little or no training in the proper and effective use of these materials, meaning a reasonable possibility exists of over- or under-application as well as the potential for creation of unrecognized resistance issues. This possibility is especially true for the indiscriminate use of aerosol foggers as well as concentrated pesticides that require mixing with water prior to application. Additionally, the health and well-being of sensitive individuals (e.g., asthmatics and chemically sensitive people) and their pets (especially birds and fish) could be affected by the unexpected drift of these pesticides into their yards, open windows, and neighborhood parks.

Second, the potential exists for increased use of inappropriate or unregistered materials such as bleach, oil, gasoline, diesel fuel, etc., in an effort to deal with mosquitoes. Their use can cause significant environmental harm compared to materials applied in accordance with label requirements by trained, licensed professionals.

Third, many members of the public lack a general understanding of IPM practices and procedures. Therefore, increased mosquito-human interactions could lead to the increased use of non-IPM practices to provide rapid relief from mosquito bites as well as address any fears concerning reports in the media of increased mosquito-borne disease.

Fourth, as mentioned earlier, some mosquito-borne diseases such as West Nile virus pose a risk to native bird species, including some species of concern such as yellow-billed magpies, hawks, and owls (Crosbie et al. 2008; Fitzsimmons 2013; LaDeau et al. 2007; Nemeth et al. 2007, 2009; Sovada et al. 2008).

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