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Design standards within constructed wetlands for the reduction of mosquito populations in Clark County, Nevada

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DESIGN STANDARDS WITHIN CONSTRUCTED WETLANDS FOR THE
REDUCTION OF MOSQUITO POPULATIONS IN
CLARK COUNTY, NEVADA.

by

Philip Bondurant, REHS

Bachelor of Science
Southern Utah University
2005

A thesis submitted in partial fulfillment of
the requirements for the

Master of Public Health
Department of Environmental and Occupational Health
School of Community Health Sciences
Division of Health Sciences

Graduate College
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December 2010

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THE GRADUATE COLLEGE

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Phillip C. Bondurant

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ABSTRACT

Design Standards within Constructed Wetlands for the Reduction Mosquito Populations in Clark County, NV.

by

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Wetlands are considered one of the most productive ecosystems in the world and provide many benefits to the environment. However, the slow moving and sometimes stagnant water created by the vegetation in the wetland creates an ideal environment for the proliferation of mosquitoes. Mosquitoes are the most important insect disease vector worldwide. The presence of mosquitoes within wetlands increases the risk of disease transmission among workers and visitors creating a public health concern. Effective design standards aimed at reducing mosquito breeding habitat should be implemented during the construction and planning phase of wetland development to effectively reduce the mosquito populations. This research evaluated the presence of mosquitoes within two wetlands in Clark County, Nevada; one constructed using the Environmental Protection Agencies suggested guidelines for mosquito reduction, the other not. During the peak mosquito season (March-October), traps were set at two wetland sites on a monthly basis. This trapping occurred for the span of two mosquito seasons in the Las Vegas valley. Trapping data were evaluated to determine if one location produced a lower mosquito

population when compared to the other. It was found that the amount of mosquitoes produced between these two locations were statistically different. Furthermore, the same information was used to determine the dominant mosquito species within the wetland and then evaluate the possibility of disease transmission among this species. *Culex (Cx.) tarsalis* was the most common mosquito species from both wetlands making up 56% (2829 of 5059) of all mosquitoes captured and consequently the most important mosquito-borne disease vector in Clark County. However, 97% of the *Cx. tarsalis* samples originated from site one (2741 vs. 88). The results of this study showed that wetland location two, constructed using EPA supported guidelines and through the implementation of these designs, limited the overall mosquito population, thereby reducing the potential for disease transmission among known disease vectors within Clark County, NV.

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DEDICATION

Dedicated to my greatest achievement in life, my family. I want to give special thanks to my wife, Jolie. She is my best friend. Thank you for waiting up on nights I came home late from class, warming up leftover dinners, and supporting me when I was a stressed out basket case. You deserve as much recognition for the hard work behind this degree as I do. Thank you.

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CHAPTER 1

INTRODUCTION

Wetlands are considered one of the most productive ecosystems in the world. They are home to innumerable species of microbes, plants, mammals, reptiles, insects, birds and amphibians. The biodiversity of wetlands rivals that of rain forests and coral reefs (EPA, 2005). Wetlands, both constructed and natural, greatly impact the environment in a positive manner. With far reaching benefits, popularity of artificial wetlands is growing dramatically. With the many crucial environmental and socio- economic functions provided by wetlands, their existence is vital, and the presence of healthy wetlands should be encouraged. During the planning of the wetland, an Integrated Mosquito Management approach is suggested for the reduction of mosquito populations within the wetland (Knight et al., 2003). By seeking out those changes to reduce and eliminate mosquito breeding sources, the potential public health hazard posed by mosquitoes can be limited.

The name wetlands is a collective term used to describe the various types of marshes, swamps, bogs, and fens that meet the hydrology standards given to a wetland. Throughout history, much of the wetlands areas in the United States have been drained and converted to farmland, filled for housing developments and industrial facilities, and used as receptacles for waste (Yuhua, 1996; USGS, 1997). These human activities continue to adversely affect, destroy, or limit the function of wetland ecosystems, consequently impacting the environment. Knowledge of the benefits provided by wetlands has fueled the effort to restore lost wetlands and has aided in the popularity of wetlands construction.

Artificial wetlands are created to take advantage of the many benefits provided by a functioning wetland. As a natural, low cost method for water filtration and purification, many states are creating wetlands to help with available water resources. Success stories exemplifying the effectiveness of wetlands can be found throughout the United States. The State of South Carolina uses the Congaree Bottomland Hardwood Swamp for purification of watershed. It is estimated that the Congaree swamp effectively removes the same volume of pollutants that a \$5 million treatment plant would eliminate (EPA, 2006). The cost to operate an artificial wetland is significantly less when compared to a standard power operated plant.

Although the attractiveness for this type of wetland system continues to grow, a primary concern for any wetland is the presence of mosquitoes. This concern is derived from the biting female mosquitoes within the wetlands and the possibility of disease transmission among workers and visitors. Public health officials argue that wetlands in an urban environment have the potential to increase mosquito populations, therefore increasing the chance for disease transmission (Chase, 2003). These diseases bring to light the public health implications behind the relationship of mosquitoes and wetlands, which raise questions of the benefit- cost ratio of a constructed wetland. Mosquito management plans often conflict with objectives of constructed wetlands and tend to discount the health concerns posed by mosquitoes. The combination of high mosquito populations and animals, such as birds, with the potential to carry and transmit disease to the biting mosquito population raises concern for the health and safety of the wetlands (Russell, 1998).

The setting and function of a wetland is an ideal environment for mosquitoes. In fact, mosquitoes are a large part of any healthy wetland ecosystem. Slow moving and stagnant water created by vegetation, highly organic water to support larval growth, resting sanctuaries, and available blood meals for adult females create an ideal environment for the proliferation of mosquitoes. Constructed wetlands are no different. These artificial wetland systems are constructed to mimic a natural wetland in both appearance and function (EPA, 2005). With this approach, those vectors for disease also accompany the artificial wetlands which have proven to breed mosquitoes in the same fashion as a natural wetland.

Mosquitoes are considered the most important insect disease vector worldwide (CDC, 2003). They are endemic to every corner of the globe and create an enormous strain due to economic and health costs. Mosquitoes function as obligate intermediate hosts for diseases like arboviral encephalitides (including West Nile Virus), malaria, dengue fever, chikungunya, and yellow fever, to highlight a few. Since 2004, West Nile Virus has been present in Clark County. Mosquitoes trapped at local wetlands have been tested and confirmed positive for West Nile Virus. In 2009, 256 mosquitoes in Clark County tested positive for West Nile Virus (SNHD, 2009). Although not all mosquitoes trapped from the wetlands in Clark County were positive for West Nile Virus, it verifies the presence of mosquitoes and highlights the chance for disease transmission. This information validates the need for supplementation to the wetland construction process with mosquito management in mind.

Significance

Recently, several wetlands have been evaluated in Clark County, Nevada for restoration or creation. These wetlands are being established to serve a variety of functions: water quality, aesthetics, or wetlands mitigation. Regardless of the reason, wetland construction is present within Clark County. In a setting like Clark County, where the landscape is primarily desert and water is in high demand, areas with water, like a wetland are likely to attract both visitors and mosquitoes. This combination of high mosquito populations, public presence, and the existence of disease could have serious public health consequences. However, if constructed in a manner that eliminates breeding sources, the opportunity for disease transmission among the general public is reduced due to acceptable mosquito populations within the area.

There is a large literature base containing guidelines for best management practices (BMP's) in mosquito control for constructed wetlands (AMCA, 2009). Typically, these reports are written specific to a geographic region, but some parallels can be found in each document. Many of the leading states, in both constructed wetlands and mosquito control, have published BMP's for wetlands specific to their geographic region. New Jersey, Maryland, Florida, California, and Utah are a few states that have recognized the need for such material and have published specific criteria for the construction of wetlands. Each document contains specific elements designed to meet the needs of each geographical area. No such document exists in Clark County, Nevada. This study looks to address the BMP's for constructed wetlands within the county during construction or renovation of wetlands. Furthermore, information is provided for the establishment of

mosquito monitoring plans, vegetation management, and mosquito management to provide insight for mosquito reduction after completion of the project.

Goal

The goal of this study is to establish a living document that can be used for reference during the construction of a wetland to assist in the decision making process. It will aid in the promotion of a healthy wetland, which naturally limits mosquito populations within the wetland. Low mosquito counts are vital for reducing the public health threat for mosquito borne disease transmission and nuisance complaints. It is evident wetlands are necessary for the success of the environment and are growing in popularity across the country. This document looks to address the public health concern and provide solutions for mosquitoes within the wetlands.

It is believed, by suggesting effective precautions for those involved in the design of the wetland, choices can be made with mosquito control in mind. Help identifying those situations that may create breeding sources are addressed and a suitable alternative is provided to avoid problems from the beginning. With implementation of these suggestions during the early stages of development, wetlands staff and management can effectively reduce and manage mosquito breeding from the day of inception. The presence of a healthy, sustainable wetland will effectively minimize mosquito breeding while maintaining its environmental and economic benefits.

Objective

This research looks to address the following objectives:

1. To develop a document that will be used by the local Vector Control agency to provide recommendations for the reduction of mosquito populations through design standards to project planners and construction managers during the construction phase of wetlands in Clark County, Nevada.
2. Evaluate mosquito populations in two separate wetlands with varying degrees of mosquito control to confirm that design standards effectively reduce mosquito populations.

Hypotheses

Hypothesis One: Two wetland locations exist within Clark County, both of which vary greatly in construction. Location one is a recreational wetland which promotes the growth of vegetation. Location two is a waste water treatment plant designed to limit or control vegetation using EPA guidelines. Through the implementation of design standards proven to reduce vegetation, thereby eliminating mosquito breeding habitat, location two will produce less mosquitoes than location one.

Hypothesis Two: By analyzing historical trapping data from both sites, it will be determined that the population of one mosquito species is more abundant than other species in the wetlands of Clark County, Nevada.

CHAPTER 2

WETLANDS

Wetlands are fragile ecosystems that provide many crucial services to the environment. Under Federal regulation, Section 404 of the Clean Water Act (Appendix I), a wetland is defined as:

“Those areas that are inundated or saturated by surface or ground water (hydrology) at a frequency and duration sufficient to support, and that under normal circumstances do support, a prevalence of vegetation (hydrophytes) typically adapted for life in saturated soil conditions (hydric soils). Wetlands generally include swamps, marshes, bogs, and similar areas (40 CFR 232.2(r)).”

Wetlands are regulated by the U.S. Army Corps of Engineers (Corps) under Section 404. According to the Corps, for an area to be considered a wetland under their jurisdiction, it must demonstrate all three characteristics: hydrology, hydrophytes, and hydric soils (US ACOE, 1987). It is important to note that natural areas that function as a wetland in the environment, but do not exhibit all three characteristics described above, do not qualify under the regulatory power of the Corps. Therefore, activities in these wetlands are not regulated under the Section 404 program (EPA, 2006).

The U.S. Fish and Wildlife Service define a wetland as:

“lands that are transitional between terrestrial and aquatic systems where the water table is usually at or near the surface or the land is covered by shallow water, and that have one or more of the following attributes:

1. At least periodically, the land supports predominantly hydrophytes;
2. The substrate is predominantly undrained hydric soil; and,
3. The substrate is non-soil and is saturated with water or covered by shallow water at some time during the growing season of each year.”

Wetlands are found in a variety of biomes throughout the world. Locations and type differ from flat vegetated areas, landscape depressions, and between aquatic and terrestrial ecosystems including the edges of streams, rivers, lakes, washes, and coastlines. Inland wetlands receive water from precipitation, ground water and/or surface water. Coastal and estuarine wetlands receive water from precipitation, surface water, tides, and/or ground water (Mitsch, 1993).

The type of soil, vegetation, and animal communities present in a wetland is determined by the level of saturation. Each wetland may support both aquatic and terrestrial species specially adapted to the individual characteristics of wetland soils (Cowardin, 1979). Although each wetland is unique and different, the hydrology, soil, and vegetation are the key characteristics of a wetland.

Constructed wetlands mirror the example created by natural wetlands. A large basin is created with the intent to hold water, a form of substrate, and vascular plants to aid in the purification of water. It should be understood these components can be manipulated to prevent mosquito breeding (EPA, 1998). By definition the creation of a wetland, regardless of its purpose, is defined by Mitsch and Gosselink as:

“The conversion of a persistent upland or shallow water area into a wetland community by human activity”

Although the definition of a wetland provided by federal agencies, states, text book authors, and scholars will vary, for the scope of this paper, wetlands are lands on which water covers the soil or is present either at or near the surface of the soil or within the root zone, all year or for varying periods of time during the year, including during the growing season. The recurrent or prolonged presence of water (hydrology) at or near the soil surface is the dominant factor determining the nature of soil development and the types of plant and animal communities living in the soil and on its surface (Phytoremediation & Constructed Wetlands, 2008). Wetlands can be identified by the presence of those plants (hydrophytes) that are adapted to life in the soils that form under flooded or saturated conditions (hydric soils) characteristic of wetlands. There also are wetlands that lack hydric soils and hydrophytic vegetation, but support other organisms indicative of recurrent saturation (Mitsch and Gosselink, 1993). All wetlands, regardless of its nature, have one characteristic in common: the hydrologic condition of the soil presents water at the surface, or near the surface, periodically.

Necessity of Wetlands

During the early 1600's the continental United States contained more than 221 million acres of natural wetlands. Throughout the last 400 years, wetland acreage has been reduced to 103 million acres. Six states lost as much as 85% of the natural wetlands during this time frame (USGS, 1997; Figure 1). With more than half of the wetlands area gone, the United States Government decided the remaining wetlands needed protection.

Percentage of Wetlands Acreage Lost, 1780's-1980's

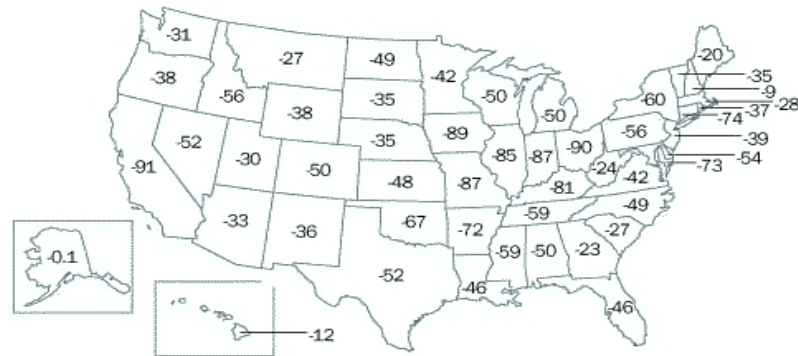


Figure 1. Twenty two states have lost at least 50% of their naturally occurring wetlands. Nevada lost 52% of its wetlands during this time frame. Mitch and Gosselink Wetlands. 2nd Edition. Van Nostrand Reinhold. 1993.

Due to their sensitive nature and rapid loss, wetlands are now regulated under the Clean Water Act (CWA) Section 404 (Appendix I) by the EPA and Corps. Under this act, wetlands may not be altered, created, or destroyed without the consent of the EPA or Corps (US ACOE, 1987).

Until recently, wetlands were drained to accommodate urban development, agriculture, and flood control. With a new understanding of the provided benefits, as well as regulations governing mitigation, wetland restoration and creation is happening country wide and in Clark County, Nevada. The wetland systems, be it artificial or natural, provide many benefits to the environment as well as local economies.

Wetland Benefits

Water quality: Clean drinking water is a finite resource. Natural methods to purify water are effective, but not efficient enough to meet the demands for clean water. Wetlands are known for their ability to capture sediments and filter pollutants. As water flows through a wetland, suspended solids are trapped or settle out. Pollutants, such as fertilizers, are broken down by biological processes to a less soluble form that is inactive or can be absorbed by plants. In both cases, the result is cleaner, usable water. This idea is being implemented to treat municipal runoff. These wetlands are being constructed at a fraction of the construction cost and operation budget of a conventional system with the same outcome (EPA, 2005; Figure 2).

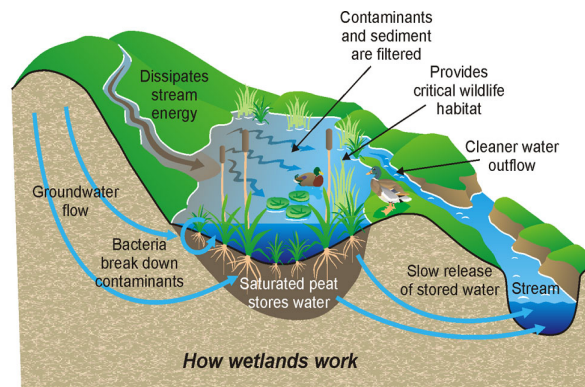


Figure 2. Overview of how wetlands function.
Image from <http://geopanorama.rncan.gc.ca>

Support of wildlife: A variety of wildlife depend on the presence of wetlands. Although healthy wetlands will support a large range of species, bird populations seem to be the greatest beneficiary. Eighty percent of America's breeding bird population and almost half of the 800 federally protected migratory birds rely on wetlands. As with any productive ecosystem, a diverse population of other animals is present. Reptiles and

amphibians are common wetland residents. Nearly 200 species of amphibians in North America require wetlands for reproduction. Mammals, such as beavers and muskrats, also benefit from the ideal habitat created by a wetland setting (EPA, 2006; USFWS, 2009).

Biological Activity: The success of wetlands in regard to water quality can also be attributed to the high concentration of organic matter. This matter serves the wetlands in multiple ways. First, it is the basis for the food chain within the wetlands. Smaller organisms feed on the nutrients, who in turn, feed larger organisms. This cycle continues as it travels up the food chain. Second, these nutrients make their way into nearby water systems providing nutrients, thereby increasing the productivity of the system and sustaining it for human activity, such as commercial fishing (EPA, 2006).

Biodiversity: In addition to supporting the life processes of wildlife, wetlands are also home to more than 500 endangered plant species, many of which are unique to each individual wetland. The number of actual plant species in wetlands worldwide is nearly impossible to calculate, as many have not been discovered. Ultimately, the function and purpose of wetlands depends on the diverse population of plants contained within. Both animals and plants play a vital role in the success and health of the wetlands environment (EPA, 2005).

Flood Damage and Erosion: By nature, wetlands have the ability to interrupt and slow raging waters created by a flood. Fast, dangerous currents are dissipated by vegetation as it passes through the wetlands. Torrent flows, which cause flooding, are reduced to manageable flows. This reduction in head volume will limit the chance of flooding in urban areas. Wetlands also have the ability to reduce wave potential that results in

erosion. Erosion control can be crucial for flood control and land management (EPA, 2006).

Recreation and Aesthetics: A healthy wetlands can become a destination for recreational activities. Well managed wetlands can support hunting and fishing without harming the overall output. With the large number of species present in a concentrated area, people are able to enjoy nature at its finest. Hiking, bird watching, photography, and canoeing are some activities that can be conducted in a wetlands setting. Urban wetlands are considered more visually appealing than the city sprawl. This increases the overall appearance of an area and in some cases has increased home values (EPA, 2006).

Economic benefits: It is difficult to calculate the economic value of a wetlands system. It is estimated that through the natural processes of a wetlands, \$14.9 trillion was contributed to the world's economy (EPA, 2006). In addition, recreational activities and flood control can create economic opportunities for a local economy (EPA, 2006).

Healthy Wetlands

With the large number of benefits provided by a wetland, their need is easily justified. Because this need is great, efforts should be made to sustain healthy, functioning wetlands (AMCA, 2009). By definition, a healthy wetland is one that minimizes risk to human health while maximizing the potential benefits of the wetland (EPA, 1998; SWS, 2009). A healthy wetland will sustain a biological balance, which through a natural system of checks and balances, will limit mosquito production, all while serving its intended purpose (Indiana Wetlands, 2009). Russell (1999) noted wetlands that maintain the constant presence of water, produce fewer mosquitoes, due to the diverse fauna. Functioning wetlands provide habitat for the natural predators of mosquitoes. Certain

birds, frogs, bats, fish, and insects rely on the life cycle of mosquitoes for nutrition. Therefore, the importance to preserve the natural balance in a wetland is vital for the success of the wetland and the reduction in mosquito populations.

In Essex County, Massachusetts the creation of an artificial wetland reduced urban mosquito populations by 90% (Indiana Wetlands Conservation Plan, 2009). This was accomplished through the control of floodwater, handled by the wetland, and the support of a biological balance which limited mosquito populations. When mosquito numbers are reduced the chance for disease transmission is minimized.

Unmanaged, or drought laden wetlands, actually promote disease transmission. These areas of stagnant water cannot support the level of mosquito predators to control mosquito populations as seen in healthy, well managed wetlands. In areas where drought has reduced the amount of available water, or water is available only part of the year, those natural defenses against mosquito production are not available. Therefore, mosquito larvae grow with little opposition, and mosquito outbreaks associated with disease are common (Chase and Knight, 2003).

It is evident wetlands are vital for the success of the environment and provide a lower cost means for water treatment. Mosquitoes and wetlands are synonymous with one another, for this reason, we must work to sustain healthy wetlands that function properly and reduce mosquitoes. With the suggestions in this paper, a healthy, functional wetland can be created with the goal of reducing measurable mosquito populations reduced.

CHAPTER 3

MOSQUITO ECOLOGY

Mosquitoes cause more human suffering than any other organism in history. It is estimated that malaria infects 300-500 million people worldwide each year and kills about 1 million of those individuals (CDC, 2003). In Africa, a child dies every 30 seconds from malaria. Mosquito borne diseases are not only a concern in exotic locations. Although uncommon, roughly 1300 cases of malaria are diagnosed each year in the United States (AMCA, 2005). More recent is the endemic presence of West Nile Virus in the United States. Mosquitoes not only carry diseases that afflict humans, they also transmit several diseases among canine and equine populations. Furthermore, they create a horrible nuisance in areas where mosquito control is absent. Mosquito bites cause skin irritation through an allergic reaction to the mosquito's saliva. The degree of severity will depend on each individual and their natural response to the mosquito bite.

Wetland managers and personnel should become familiar with local wetland mosquito species and their characteristics (Marin and Sonoma, 2000). A complete list of mosquito species found in Clark County is available in Appendix II. A general understanding of mosquito ecology will prove valuable during the construction and maintenance of the wetland. This knowledge will aid in the decision making process, and to identify breeding sources in the future. The pesticides used for mosquito control are engineered to work during certain stages of the mosquito life cycle. Having an understanding of the mosquito life cycle and how the chemicals work will help field staff and wetland managers effectively apply pesticides. This will limit the impact of pesticides on the wetland environment and will provide the best control results. The ability to prevent,

identify, and treat mosquito breeding habitat will minimize concerns from inception of the wetlands.

It should be noted, many of the mosquito genera begin with the same letter, for instance *Culex* (Cx.) spp. and *Culiseta* (Cu) spp. For this reason, a two letter abbreviation is used to identify individual genera, which helps to avoid confusion among species. As each species is introduced for the first time throughout this paper, the genera specific two letter abbreviation will follow in parenthesis.

Life Cycle

The following review of the mosquito lifecycle is taken from the American Mosquito Control, 2005 (Figure 3):

“The mosquito goes through four separate and distinct stages of its life cycle: Egg, Larva, Pupa, and Adult. Each of these stages can be easily recognized by its special appearance.”

“Egg: Eggs are laid one at a time or attached together to form "rafts." They float on the surface of the water. In the case of *Culex* (Cx.) and *Culiseta* (Cu.) species, the eggs are stuck together in rafts of up to 200. *Anopheles* (An), *Ochlerotatus* (Oc.) and *Aedes* (Ae.), as well as many other genera, do not make egg rafts, but lay their eggs singly. *Culex*, *Culiseta*, and *Anopheles* lay their eggs on the water surface while many *Aedes* and *Ochlerotatus* lay their eggs on damp soil that will be flooded by water. Most eggs hatch into larvae within 48 hours; others might withstand subzero winters before hatching. Water is a necessary part of their habitat.”

“Larva: The larva (plural - larvae) lives in the water and comes to the surface to breathe. Often time they are referred to as “wigglers” or ‘wigglers’. Larvae shed (molt)

their skins four times, growing larger after each molt. Most larvae have siphon tubes for breathing and hang upside down from the water surface. *Anopheles* larvae do not have a siphon and lie parallel to the water surface to get a supply of oxygen through a breathing opening. The larvae feed on microorganisms and organic matter in the water. During the fourth molt the larva changes into a pupa.”

“Pupa: The pupal stage is a resting, non-feeding stage of development, but pupae are mobile, responding to light changes and moving (tumble) with a flip of their tails towards the bottom or protective areas. This is the time the mosquito changes into an adult. This process is similar to the metamorphosis seen in butterflies when the butterfly develops- while in the cocoon stage- from a caterpillar into an adult butterfly. In *Culex* species in the southern United States this change can occur in two days during the summer. When development is complete, the pupal skin splits and the adult mosquito emerges.”

“Adult: The newly emerged adult rests on the surface of the water for a short time to allow itself to dry and all its body parts to harden. The wings have to spread out and dry properly before it can fly. Blood feeding and mating does not occur for a couple of days after the adults emerge.”

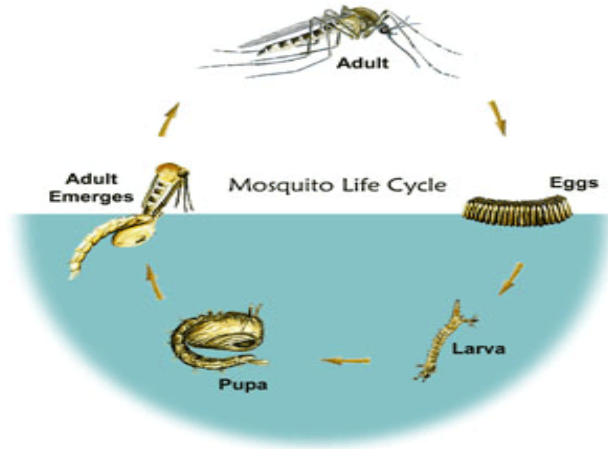


Figure 3. Mosquito life cycle. Image from AMCA. www.mosquito.org

The length of each stage is temperature dependent. *Culex tarsalis*, the primary vector for West Nile Virus in Clark County, may complete its life cycle in 14 days at 70° F. During the peak mosquito season, when temperatures consistently reach 100+° F, the life cycle may be completed in 5 days. Other species have developed even shorter life cycles. *Aedes vexans*, commonly named the flood water mosquito, is present in rural Clark County where irrigation practices mimic flood conditions. When excess irrigation is present, eggs planted in the soil hatch viable larvae. Floodwater mosquitoes can develop from egg to adult in as little as 72 hours.

Flight Range

Most species have flight ranges of 1-3 miles. Other species have been found to migrate 100 miles from known breeding sources under exceptional circumstances. *C. tarsalis*, the primary vector for West Nile Virus in Clark County, has flight range of 1-2

miles (VMCA, 2009). During windy conditions, mosquitoes can become caught in updrafts that lead them to prevailing winds which can carry them great distances.

In Clark County, all of the current manmade wetlands are within the flight range of housing developments for *C. tarsalis*. With large communities in range of mosquitoes, it is important to implement all possible mosquito reduction measures to keep the mosquito population at a minimum. In doing so, nearby residents will not be concerned with adult mosquitoes from a nuisance or disease transmission standpoint.

Habitat

Mosquito larvae occupy various habitats in a wide range of environmental conditions. Each species will have certain factors that draw them to specific habitats. Mosquitoes are associated with water where their young can develop. For water to be conducive to mosquito breeding, it is important the water remains standing long enough for the larvae to fully develop. Mosquito larvae flourish in shallow, standing water, with low oxygen content, and a highly organic content. Mosquitoes can breed wherever water collects, including wetlands, abandoned swimming pools, and storm drains. The pH of the water can have little effect as some species inhabit waters, either acidic or basic, that record at either end of the pH scale (AMCA, 2009).

Healthy wetlands do not promote ideal mosquito breeding habitat (Indiana Wetlands, 2009). That is why it is important to establish guidelines such as this document that are specific to geographical areas. An understanding of mosquito habitat during the construction phase will minimize the need for biological controls during the maintenance phase of the wetland. Effective control measures that are implemented during

construction will create a healthy environment for wetlands habitat without large scale mosquito concerns.

Mosquito Borne Disease and Transmission

Mosquitoes act as vectors in the transmission of disease. Most commonly known for the spread of malaria, mosquitoes also transmit several other diseases that cause harm to people. The most recent mosquito borne epidemic in the United States started in 1999 with the discovery of West Nile Virus fever. According to the American Mosquito Control Association, “The introduction and spread of West Nile virus in the United States has reawakened an appreciation of mosquitoes as vectors of diseases for mosquito-borne diseases were once quite prevalent in the United States and, indeed, played a major part in shaping our nation's destiny.” Mosquito borne disease outbreaks have been found in U.S. history as far back as 1780 when Dr. Benjamin Rush first described dengue fever in Philadelphia and 125,566 cases of malaria were reported in the U.S. as recent as 1934 (AMCA, 2005). These diseases are no longer endemic to the United States. Mosquito control agencies in conjunction with public health intervention have worked diligently to create a relatively disease free society.

West Nile Virus (WNV) was first discovered in the United States in New York during the summer of 1999. The virus quickly spread west and was first detected in Clark County in 2004 (SNHD, 2005). At least 60 species of mosquitoes have been found infected with the WNV in the United States with 13 of those species found in Clark County. WNV has remained constant in Clark County, with surveillance efforts continuing to discover positive mosquitoes and human cases. In 2009, 11,337 mosquitoes were trapped in Clark County with 256 mosquitoes testing positive (2%) and 12 human

cases being reported (SNHD, 2009). Although the risk of infection for WNV is relatively low, it is evident disease transmission among mosquito populations in Clark County is occurring.

Aside from disease, mosquitoes also create a nuisance concern. Outdoor events can be severely hindered when mosquitoes are present. In an area like Clark County, where the landscape is dry and arid, water and moisture come at a premium. Therefore, those areas used by people to retreat from the heat are also favored by the local mosquitoes. This is a cause for concern as the number of mosquitoes and people become concentrated in areas around water, the chance for exposure increases. In an urban wetland, the flight range of a mosquito will easily cover the distance between the wetland habitat and the nearby housing development. This will indefinitely increase the nuisance complaints and drive down property values (EPA, 2006).

Mosquito borne disease transmission occurs when a female mosquito, acting as a vector for disease, pierces the skin of the unsuspecting host in search for a bloodmeal (Figure 4). As the female mosquito inserts her proboscis, the virus laden saliva is allowed to enter the host through the newly created break in the skin (Dept. of Medical Entomology, 2009). From there, depending on the dynamics of the disease, replication and the effects of infection may shortly follow. This mechanism within the mosquito is part of the natural lifecycle and necessary for the propagation of the species.

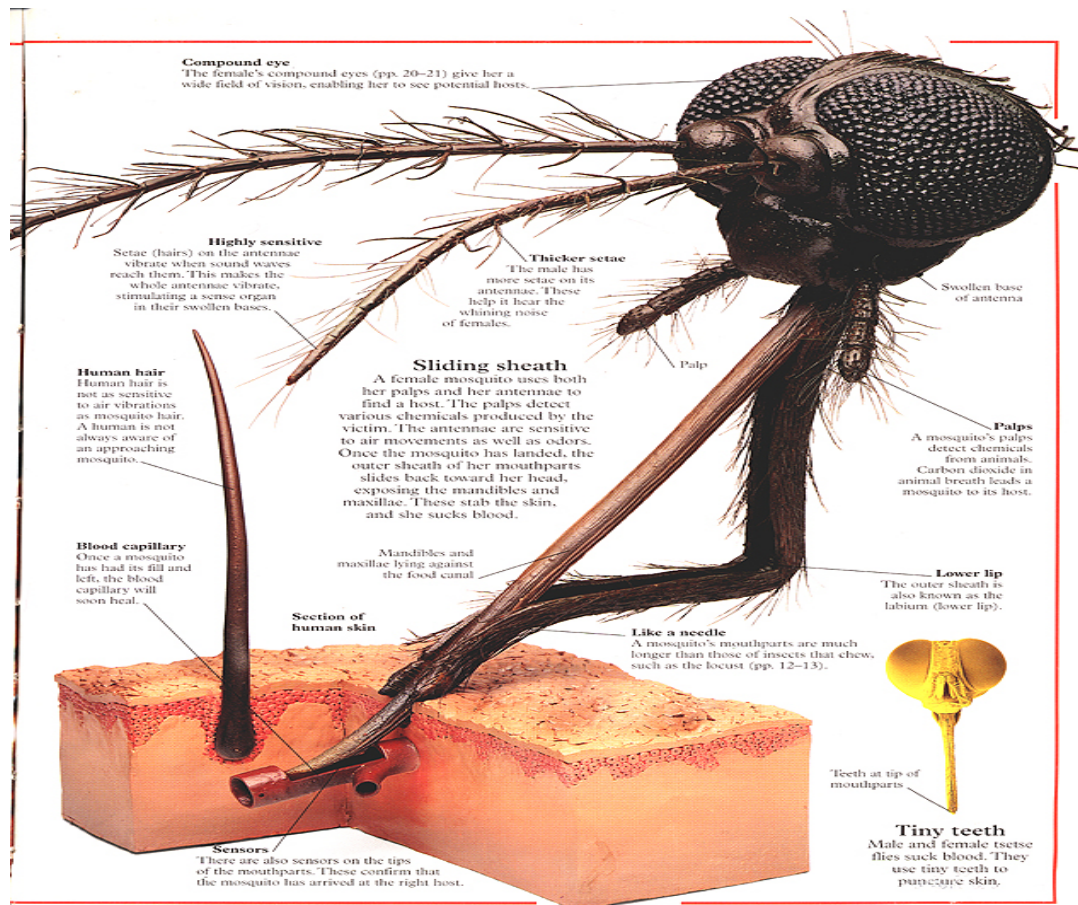


Figure 4. Method of obtaining a bloodmeal by female mosquito. Image from <http://www.bugs.org/GalleryPages/Mosquito>

This information should provide evidence that mosquito control in constructed wetlands is necessary. All efforts should be made to ensure mosquito habitat is limited during construction. Management efforts post- construction should be focused on control of the mosquitoes within the wetland. New science has made great strides in defining the transmission dynamics of mosquito borne disease, but people are still being infected. The primary concern in disease transmission is the control of the female mosquito. However, gender specific controls do not prove any more effective than a complete control approach. It is evident the need for control will be required, therefore should not be

overlooked in wetland management plans. The following few sections provide a background on mosquito control and describe various methods of mosquito control.

Mosquito Control

The objective of mosquito control is to reduce contact between mosquitoes and humans. This can be accomplished through a combination of three types of control: Physical, Biological, and Chemical. However, the most important aspect of mosquito control is surveillance (Knight et al., 2003; Russell, 1998; Walton, 2003). The results of surveillance efforts within the wetland are what should drive the type of control method employed. Only those measures which will be most effective should be used. This will limit the impact on the environment as well as the biological balance within the wetland. Methods for surveillance and development of mosquito control plans will be discussed in Chapter 6.

Successful mosquito control programs rely upon principles that exploit the mosquito's vulnerabilities. Complete eradication of mosquitoes is impossible and even in healthy wetlands, mosquito production should be expected. Since not all mosquito species found in Clark County transmit disease, control measures for mosquitoes will vary depending on the habitat (SNHD, 2008). Therefore, the goal of mosquito control efforts is to maintain acceptable levels of mosquitoes through effective control methods. A general knowledge of the target mosquito vector will help with the allocation of control resources. The proper combination of physical, biological, and chemical methods to control mosquitoes will prove more effective than the use of one method alone (EPA, 2009). Mosquito control will be an important part of manmade wetlands and wetland

management. Therefore, wetland management should understand the ways in which mosquito populations can be controlled.

Physical Controls

Physical methods of mosquito control are the most effective and provide long term benefits (Society of Wetland Scientists, 2009). This method refers to physically altering the environment or landscape to limit habitat. This term is also known as source reduction. Ultimately, the physical change in the landscape will make the site less suitable for mosquito production. Physical control can be accomplished in a variety of methods. The overall goal of source reduction is to remove the element that is promoting the production of mosquitoes. Vegetation removal, draining, trenching, grading, and diverting are some means of source reduction. Methods of source reduction through vegetation management are discussed later in the document.

Site design and pre-planning are very important in determining the need for source reduction. During the design stage, if those areas within the wetland that pose a problem can be identified, then changed or removed, the need for physical control will be limited. Furthermore, by understanding mosquito ecology, wetlands can be engineered to reduce mosquito populations and the need for physical control will be minimal.

Biological Controls

Biological control involves augmentation of natural predator species to manage mosquito populations (EPA 2009). There are several methods of biological control. *Gambusia affinis*, or mosquito fish are the most well known. Mosquito fish will consume large numbers of mosquito larvae, upwards of 500 a day, effectively limiting mosquito

populations and creating generational gaps (San Mateo County, 2008)). Other fish have been known to feed on mosquito larvae as well, but not at the same capacity as *G. affinis*.

Other effective biological controls include *Bacillus thuringiensis* (BT). BT consists of the dead spores from the natural soil bacterium *B. thuringiensis*. *Bacillus thuringiensis israelensis* (BTI) is a specific species of BT that has proven very effective in controlling mosquitoes by interfering with the digestive system of the larvae. When the BTI spores are eaten by the mosquito larvae, they damage the gut cells and quickly paralyze them, which cause the larvae to drown. It is very selective in that it affects mosquito larvae without harming the other inhabitants of the wetland. BTI is usually bound in different materials, such as corn granules, and applied by hand or dropped by helicopter in large areas (Figure 5). The spores are then released once the product hits the water, where the larvae can consume the spore. BTI is only effective on the larval stage of mosquito development (EPA, 2007). Although BTI is a biological control agent, it is sometimes grouped into the chemical aspect of mosquito control by those who do not completely understand how it works.



Figure 5. Common *BTi* form.
Corn cob granules infused with
BTi product.

Some other biological control methods have proved successful, but to a lesser degree. Dragonfly nymphs and adults will consume mosquito larvae in breeding waters, and adult dragonflies, will eat adult mosquitoes. Certain insects, crustaceans, copepods, nematodes, and fungi, all of which are natural to wetlands, have been known to affect the development of mosquito larvae (AMCA, 2005). Some public agencies use other predators such as birds, bats, lizards and frogs, but evidence supporting the effectiveness of each is scarce (EPA, 2007).

Chemical Controls

The chemical control of mosquitoes refers to the use of pesticides. Pesticides are used to control the larval stage (larvicides), pupal stage (pupicides), and adult mosquitoes (adulticides). The application of mosquito specific pesticides should be verified through the presence of the target mosquito stage, as demonstrated by surveillance efforts. The act of blindly applying chemicals without evidence confirming a need for application for need is prohibited (EPA, 2009). Furthermore, using chemicals in this manner is ineffective and a waste of control resources. For this reason, all chemical control methods should be based on scientific evidence and driven by surveillance.

Best management practices (BMP's) endorsed by EPA and the Centers for Disease Control (CDC) recommend the application of larvicides and adulticides when surveillance indicates that physical and biological control measures have proven inadequate to prevent imminent disease outbreaks. The State of Nevada requires any personnel handling restricted use pesticides to be certified operators trained in the special handling requirements of these chemicals. This ensures mosquito control products are applied at the suggested rates and in a safe manner, ultimately minimizing any damage to the environment.

The most efficient way to control mosquito populations is through larval control. Larvicides utilize insecticides targeted at immature mosquitoes. They are engineered to inhibit mosquito development and safely counter each stage of the mosquito life cycle. The intention of larvicides is to control the immature stages at the breeding source before they are allowed to disperse into the environment as biting adult populations. In doing so, generational gaps are created and the risk of arbovirus transmission is minimal. Larviciding is more effective and target-specific than adulticiding, but less permanent than source reduction (EPA, 2009). An effective larviciding program is an integral part of any integrated mosquito control operation and will reduce, if not eliminate, the need for adulticiding applications.

Larvicides are applied directly to the water where the greatest concentrations of larvae exist. Because they are used in sensitive environments, the application rate for each larvicide is calculated on the basis of its toxicity profile and degradation characteristics. Formulations will be labeled specifically for larviciding and will describe habitats where they are effective. The application of a larvicide(s) should be point specific to validated

larval locations confirmed through surveillance efforts. As a result, there is less impact on the environment and resources are not wasted (EPA, 2007). Before purchasing or applying larvicides, it is important to verify the following:

- Material sought is labeled for use on mosquitoes
- Physically control the application to the designated area
- Use the labeled amount to minimize impacts on non-target organisms
- Larvicide formulations (i.e., liquid, granular, solid) must be appropriate to the habitat being treated so the product will reach the desired area

Accuracy of application is important in minimizing environmental impact and ensuring the chemical was able to properly treat the area. If the product formulation being used is unable to reach the larvae or a relatively small area is missed, an emergence of large mosquito broods will result (Russell, 1998).

Microbial larvicides are bacteria that are registered as pesticides for control of mosquito larvae. The duration of the product is dependent on the species of mosquito, environmental conditions, product formulation, and water quality. Microbial larvicides act through the ingestion of live bacteria or a bacterial spore. The mode of action for both is the same. The toxin produced by the bacteria disrupts the gut in the mosquito by binding to receptors present in insects, but not in mammals. This makes microbial larvicides ideal for use in wetlands. BTI and *Bacillus sphaericus* (*B. sphaericus*) are the two most common microbial larvicides used. Both are naturally occurring soil bacteria registered for control of mosquito larvae (EPA, 2007). There are 26 BTI products labeled specifically for mosquito control in the United States.

Surface agents come in two varieties, monomolecular films and oils. Monomolecular films (MMF) are low-toxicity pesticides that spread across the surface of the water one molecule thick. The thin film interrupts the critical air to water interface that creates surface tension necessary for larval development. The larvae can no longer attach their siphon tubes to the surface, causing them to drown (Agnique MMF, 2006). With the surface tension removed, MMF's incidentally control pupae and adult mosquitoes. Pupae will drown, just as the larvae. Adult mosquitoes will not be able to rest on the surface of the water for oviposition. Rather they will sink in the water, eventually drowning. Films are subject to UV degradation and break down quickly. In Clark County, they are used often, but only counted on to provide control for 48-72 hours from application. When used according to label directions, MMF's pose little threat to the environment and wetlands.

Oils, like films, are pesticides used to form a layer on top of water to drown larvae and pupae. However, oils differ from MMF's by their mode of action; rather the sheet of oil becomes impenetrable and do not allow the subject mosquito to breathe oxygen through the siphon tube. Oils are derived from petroleum distillates and are used in agriculture throughout the United States in addition to controlling mosquitoes. They are also subject to accelerated degradation by UV light. Therefore, they also pose little threat to the environment when used according to the product label (AMCA, 2005).

Contact larvicides are only effective when mosquito larvae come in contact with it. Chemicals must be absorbed through the insect's chitin exterior for it to be effective. Contact larvicides are engineered to affect the nervous system of mosquito larvae. The most popular contact larvicide is Temephos. Temephos is the only organophosphate

registered with the EPA for larvicidal use (EPA, 2001). Temephos is used in areas of standing water, where organic content is extremely high and oxygen content is low, such as sewage ponds. Furthermore, it is an important resistance management tool which to prevent mosquitoes from developing resistance to the bacterial larvicides (Marin/Sonoma Mosquito Control, 2000).

Insect growth regulators (IGR's) prevent normal metamorphosis of the target insect from larvae to adult by interfering with the endocrine system and hormone levels. IGR's do not produce the nondiscriminatory, rapid toxic effects that are associated with contact larvicides. Instead IGR's maintain juvenile hormone levels in specific targets. By creating a hormonal imbalance within the larvae, the larvae cannot properly develop (Central Life Sciences, 2010).

IGR's have become popular in mosquito control programs due to the specificity of the chemical. When used within label specifications, the environmental impact of IGR's is greatly reduced and poses no risk to non-target organisms, including humans (Central Life Sciences, 2010). Methoprene is the IGR compound typically used in mosquito control. The proper use of Methoprene does not pose unreasonable risks to wildlife or the environment. Toxicity levels to birds and fish are low, and it is nontoxic to bees.

Pupicides act in the same manner as most larvicides. In fact, most larvicides are labeled to treat the pupal stage as well. However, any larvicide that has to be ingested (i.e. microbial larvicides) will not work. Pupae do not eat; therefore ingestion of the necessary particle is impossible. Some evidence suggests that Methoprene products have some effects on the molting process, but the data are inconclusive.

Most pupicides work by drowning or suffocating the pupae. Pupal control is usually accomplished with the application of any MMF or oil and categorized along with larval control.

When mosquito problems necessitate the use of insecticides, generally it is best to employ larvicides and pupicides. However, if a reduction in adult mosquito populations is not occurring through larviciding, an adulticide spray should be considered. Adulticides are pesticides designed to kill adult mosquitoes. For an integrated approach in the management of mosquito populations, the ability to control adult mosquitoes is necessary. Adulticides, when used appropriately, will have an immediate impact to reduce the number of adult mosquitoes in an area. This reduction in numbers can be used to combat an outbreak of mosquito-borne disease or reduce a nuisance infestation of mosquitoes in a community.

Mosquito adulticides are applied as ultra-low volume (ULV) sprays with extremely small droplet sizes ranging from 10-20 microns (EPA, 2009). ULV sprayers are designed to dispense micron sized droplets that stay aloft for extended periods of time. By increasing the amount of time in the air, the droplets have a higher chance of contacting the female mosquito and causing death. ULV applications magnify pesticides, allowing for small amounts of pesticides to treat large areas. The small amount used minimizes exposure and risks to people and the environment. Depending on the product, $\frac{3}{4}$ ounce to 3 oz per acre can be used with great results. The EPA has determined that insecticides labeled for adulticiding, when used within the bounds of the label, do not pose unreasonable risks to humans, wildlife, or the environment (EPA 2009; AMCA 2009).

Adulticides in the United States fall into two chemical categories, organophosphates and pyrethroids. Organophosphates work by blocking necessary enzymes in nerve endings that transmit informational signals, essentially causing death (IDPH, 2009). Malathion and Naled are the only two organophosphates currently used for adult mosquito control in the U.S. (EPA, 2007). Malathion has become a popular choice among mosquito control districts due to its low price, proven efficacy and toxicity levels equal to table salt.

There are currently four pyrethroid products on the market, pyrethrins, resmethrin, sumethrin, and permethrin. These products also work through blocking essential enzymes necessary for nerve transduction. All of these products are produced from chrysanthemum extract. These synthetic derivatives are 50 times less toxic than the natural insecticides, while proving to have the same efficacy.

There is a large body of scientific literature demonstrating significantly reduced trap counts after adulticide applications (Knight et al., 2003). However, adulticides are not selective and many times reduce population counts of beneficial insects as well. Furthermore, evidence of chemical resistance in mosquitoes is higher among adulticides than larvicides (Strong et al., 2008). Adulticide applications should not be the sole means of control in an urban setting. Mosquito control should utilize all approved means to reduce populations below transmission threshold. All insecticide selections, most importantly adulticides, should be based on the timing of the application, distribution and behavior of the target mosquito species, temperature, and time of year. This will improve the effectiveness of the chemical and minimize environmental impacts.

A complete list of EPA approved mosquito control pesticides can be found in Appendix III.

Integrated Pest Management

The EPA defines Integrated Pest Management (IPM) as:

“Integrated Pest Management (IPM) is an effective and environmentally sensitive approach to pest management that relies on a combination of common-sense practices. IPM programs use current, comprehensive information on the life cycles of pests and their interaction with the environment. This information, in combination with available pest control methods, is used to manage pest damage by the most economical means, and with the least possible hazard to people, property, and the environment.”

“The IPM approach can be applied to both agricultural and non-agricultural settings, such as the home, garden, and workplace. IPM takes advantage of all appropriate pest management options including, but not limited to, the judicious use of pesticides. In contrast, organic food production applies many of the same concepts as IPM but limits the use of pesticides to those that are produced from natural sources, as opposed to synthetic chemicals.”

The field of mosquito control takes the idea of IPM a step further with the implementation of Integrated Mosquito Management (IMM). IMM refers to strategies used by area control districts that are endorsed by the CDC and EPA and considered by both to be environmentally sound practices (AMCA, 2009). The outline contained in an IMM plan is specifically tailored to effectively counter each stage of the mosquito life cycle.

IMM plans involve three aspects of mosquito control and combine them to create one sensible, responsible plan. These strategies include physical control, biological control, and chemical control. IMM strategies for source reduction and the enhancement of biological control are employed in conjunction with mosquito specific larvicides and adulticides, to create a diversified plan selectively created for the control of mosquitoes (AMCA 2009; EPA 2009).

IMM plans follow a standard progression of mosquito control starting with the control of larvae. All IMM plans within Clark County, NV are aimed at the control of larval populations through water management and source reduction (SNHD, 2009). When source reduction is not a viable option, the use of the environmentally friendly EPA-approved larvicides will be used to control larval populations.

If larval control measures prove inadequate, or in the case of imminent disease, the EPA and CDC have emphasized the need for pesticides aimed at adult mosquitoes, also known as adulticides. These chemicals are applied under strict guidelines by certified applicators trained in the special handling characteristics of these products (NDOA, 2010).

The implementation of an IMM approach is vital in the success of the wetland. Every managed wetland should utilize all angles of mosquito control to ensure control while minimizing the effects on the environment. The creation of an IMM plan can be difficult and somewhat daunting. Chapter six of this document briefly details essential components of a plan, which should give a starting point. However, it is recommended that the local mosquito abatement district be contacted for their input, which will be specific to that geographic region.

CHAPTER 4

DESIGN CRITERIA

The information contained in this document will provide suggestions for manipulations to the design of artificial wetlands. These ideas are intended to reduce mosquito breeding without sacrificing the performance of the wetland. Little mention is made of funding, construction cost, topography, hydrology, and location as it is assumed this aspect of the project has been established. All permits, regulations, and legal guidance should be researched prior to construction. This document looks to provide insight for the development of artificial wetlands and act as an aid for project managers with the goal to reduce mosquitoes within the wetland. All of the suggestions within this paper, especially this section, follow guidelines set by the EPA and subsequent laws governing wetlands and wetland protection.

Many times the overall design features of a wetland conflict with the ideals of integrated mosquito management (Russell, 1998; Marin/ Sonoma Mosquito Control, 2000). The principal goal for artificial wetland design is to maximize treatment efficiency while minimizing the impact of mosquitoes (Walton, 2003). However, those processes which prove advantageous for water quality tend to be the same characteristics opposed for the control of mosquitoes. Therefore, a balance between function and safety should be found. With this approach, the benefits of the wetland will serve both parties.

Even with an abundance of research regarding the topic, an optimal design for constructed wetlands has not been discovered (EPA, 1998). Each wetland will vary in shape, size, and flora dependent on the landscape. Therefore, it should be known wetlands can be designed in a number of ways and still be successful. This knowledge

allows for changes in the construction of the wetland that will effectively reduce mosquito breeding without compromising the beneficial effects.

The EPA has created the “Handbook to Constructed Wetlands” which provides general guidance for the planning phase of wetland construction. These suggestions contained below (taken from the handbook) provide a rudimentary outline for considerations that should be made during the design of the wetland to help ensure success (EPA, 1998):

- Keep the design simple. Complex technological approaches often invite failure
- Design for minimal maintenance.
- Design the system to use natural energies, such as gravity flow.
- Design for the extremes of weather and climate, not the average. Storms, floods, and droughts are to be expected and planned for, not feared.
- Design the wetland with the landscape, not against it. Integrate the design with the natural topography of the site.
- Avoid over-engineering the design with rectangular basins, rigid structures and channels, and regular morphology. Mimic natural systems.
- Give the system time. Wetlands do not necessarily become functional overnight and several years may elapse before performance reaches optimal levels.

Strategies that try to short-circuit the process of system development or to over manage often fail.

- Design the system for function, not form. For instance, if initial plantings fail, but the overall function of the wetland, based on initial objectives, is intact, then the system has not failed.

All constructed wetlands consist of three common components: a basin designed to hold water, a form of substrate, and vascular plants. In addition, there are three site characteristics that will determine the type of the wetlands: topography, site ownership, and soil composition. These three attributes will determine if adequate natural flow is available for constructed wetlands. Otherwise, planning sessions must address the issue of elevation changes and deal with them accordingly (EPA, 1998).

The planning phase is crucial to the success of the wetland. Initial planning sessions should begin long before construction commences on the project. During this time, decisions will be made that will affect the overall success and productivity of the area. Therefore, all plans should be carefully considered and include alternative choices. Throughout the project planning sessions should continue to develop and amend ideas that will benefit the wetland while continuing to reduce mosquito breeding.

Planning sessions should involve a variety of representatives from local jurisdictions who have expertise in the subject matter. During this time, choices about the type, location, and function should be considered and addressed. The goal of each planning session should be geared toward the creation of a biologically functional structure.

The Marin and Sonoma County Mosquito Control district has created a questionnaire for wetland development and management. This questionnaire can be found in Appendix IV. This will ensure all aspects of mosquito control have been addressed prior to implementation of the wetland.

Location

It is stated earlier in the section, little would be said regarding the location of a wetland. Nonetheless, there are a few concerns that should be addressed during the planning sessions. Although vector borne disease is the primary concern of mosquito control, the nuisance factor cannot be underestimated. The presence of mosquitoes can make a wetland, and the surrounding area, uninhabitable (Interagency for Wetland Restoration, 2003). When placed in an urban setting, this can cause great concern and far outweigh the benefits provided by the wetland. Flight distances of wetland inhabiting mosquitoes can make it nearly impossible to find a location with enough distance to deter mosquito flight.

If it is deemed necessary a wetland must be placed in an urban setting, areas should be identified which display natural wetland tendencies and support current mosquito populations. It has been found in areas with naturally occurring wetlands and mosquito population that the addition of an artificial wetland has reduced the overall number of mosquitoes in the area through the support of natural mosquito reduction properties (Interagency for Wetland Restoration, 2003). This information should provide another incentive for designs which reduce mosquito breeding.

Water Quality Considerations

A determining factor in artificial wetland construction is the type of water treated. Typically, artificial wetlands are used to treat two types of water: untreated (sewage) and reclaimed. The quality of water will directly affect mosquito production within the wetland. A large number of studies are available that document the relationship between mosquito production and poor water quality (Knight, 2003; Walton, 2003). Untreated

water contains high levels of dissolved organic material which provide essential nutrients for mosquito larvae populations. With low oxygen concentrations and high organic concentrations, natural larval predators such as mosquito fish and dragonfly nymphs, cannot be supported (Russell, 1998; Walton, 2003; Chase and Knight, 2003). Furthermore, highly organic waters drastically reduce the effectiveness of many pesticides aimed at controlling the larval phase. The combination of nutrient availability, low predator resistance, and ineffective mosquito control efforts allow for larval growth and the possibility of explosive mosquito populations with the increased chance of disease transmission.

As noted above, many studies have documented the relationship between mosquito populations and water quality. The pre-treatment process removes much of the organic material from the untreated water. This water is generally referred to as non-potable water and used to irrigate parks and golf courses. This pretreatment step has been shown to considerably reduce mosquito production and overall mosquito numbers. The reduction in mosquitoes can be equated to the reduced organic content of the water, the sustainability of natural aquatic mosquito predators, and effectiveness of chemical pesticides used to control mosquito larvae. Although pretreatment of water before discharge into the wetland will reduce mosquito production, it does not guarantee the absence of mosquitoes (Knight, 2003).

Wetland Type

During the initial planning phase, two key decisions must be made which will determine the type of wetland: surface flow (SF) or sub-surface flow (SSF) and treated or un-treated water. An attribute table discussing positive and negative points of each can be found in appendix V. Extensive research should be conducted during the planning phase to determine the cost- benefit ratio for each type of wetland. This study will provide an in depth look at the overall picture for each type of wetland. This will allow for a decision according to the wetland type best suited for the location.

Surface Flow Wetlands

SF wetlands consist of compartmentalized basins called cells. The cells are filled with soil, peat or other substrate that will support adequate vegetation for water filtration (EPA, 2005; Knight et al., 2003). If necessary, berms may be constructed between ponds to create partitions between cells. General design standards indicate SF wetlands will have a soil bottom, emergent vegetation, and the majority of the water above ground (Figure 6). Two or more cells, depending on the size of the area and water flow demands, should be constructed in parallel to provide operational flexibility (EPA, 2005). This will allow for the draining of the ponds for maintenance or other needs. The design must include as much open water area as functionally possible. In doing so, predatory fish and wave action will naturally aid in the control of mosquitoes (Walton, 2003; Andrews and Pollard, 2008).

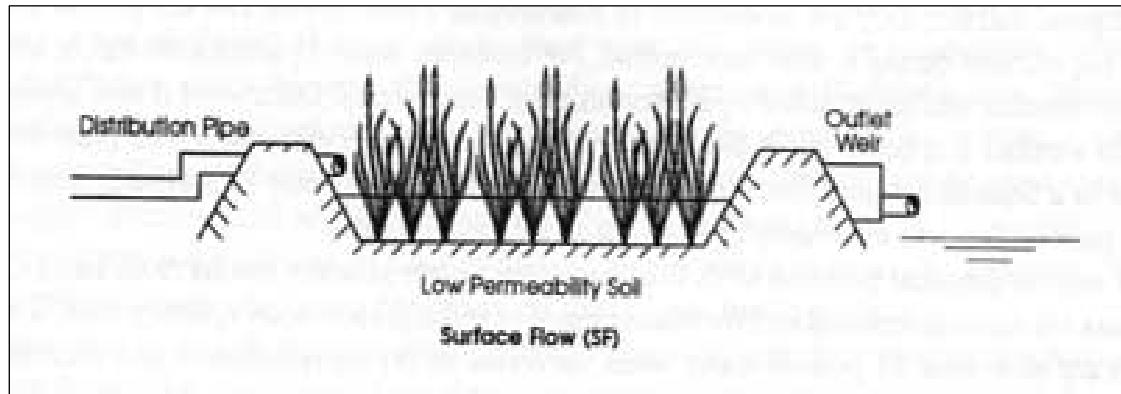


Figure 6. Illustration of surface flow (SF) wetland. Image from EPA. Handbook of Constructed Wetlands. 2005.

By design, water is filtered as it slowly makes its way through the wetland above the substrate, as previously noted. SF wetlands are densely vegetated and typically have water depths less than 1.3 ft (Kadlec and Knight, 1996). This landscape creates an ideal breeding site for mosquitoes, and should be carefully monitored.

Surface flow systems tend to cost less at startup and provide more effective means of water purification. The tendency for waterways to clog from suspended solids is far less than that of a SSF. In addition, SF wetlands provide the added benefit of wildlife habitat, including the presence of species that limit mosquito production (Kadlec and Knight, 1996; SWS, 2009; Mitch and Gosselink, 1993).

However, the maintenance required for SF wetlands far exceeds a SSF wetland. This can be seen in the amount of resources spent in managing the wetland, vegetation control, and employees (EPA, 1998; 2005). Depending on what type of wetland is created, income created by the wetland (i.e. hunting and fishing) may help to offset some of the cost associated with the operation of the wetland. Slow moving water accompanied by mosquitoes and odors are also common liabilities associated with SF systems. For those

reasons, the location of a SF wetland should be taken into consideration (SWS, 2009; Knight et al. 2003).

Subsurface Flow Wetlands

SSF wetlands are constructed in a similar manner to SF wetlands. The difference being, the cells are constructed underground using a porous material that will allow for the flow of water as well as plant growth (Knight et al. 2003; EPA, 2005). SSF wetlands are designed in one of two ways regarding flow: either horizontal or vertical. The name refers to the manner in which the water travels as it passes through the wetland. As the wetland becomes established, distinct zones are created for the improvement of wastewater (Dusel and Pawlewski, 2004).

The emergent vegetation, which is above the substrate, works to provide oxygen for biological processes below the substrate where the water purification occurs. This cycle allows the beneficial bacteria and fungi to live in the substrate as a biofilm and work to remove contaminants from the water (Dusel and Pawlewski, 2004; EPA, 1998; 2005).

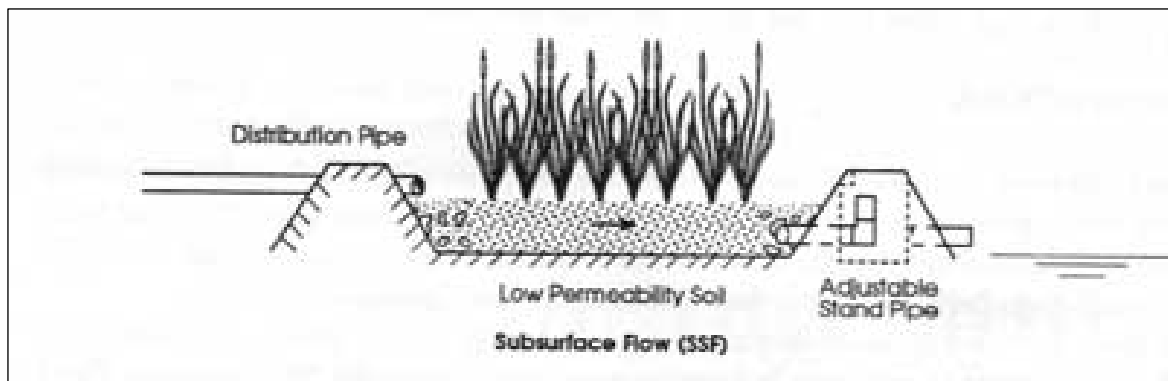


Figure 7. Illustration of sub- surface flow (SSF) wetland. Image from EPA. Handbook of Constructed Wetlands. 2005.

SSF wetlands are considered to have several advantages over the SF wetlands. Although SSF wetlands require considerably higher startup budget, the cost to maintain and run the system after completion is much lower than a SF system (Knight et al. 2003). Higher rates of contaminant removal, larger surface area promoting bacterial growth, and smaller area requirements allow SSF systems to be more productive. Other benefits include reduced odor, lower number of vectors, and no exposure risks for the public which allows for an SSF wetland to be located in urban areas (EPA, 1998).

In addition to a large construction cost, other benefits associated with wetlands are surrendered when an SSF system is used. Wildlife habitat, recreation, and some of the economic benefits linked to wetlands are no longer available to help offset reoccurring costs. Although mosquito concerns are minimal, they are not eliminated. SSF systems have a tendency to clog the filter substrate, causing water to pool outside the system. If this situation is not corrected immediately, an ideal habitat is created for mosquito larvae. This brood of mosquitoes will find little natural resistance as local populations of predatory fish are not found in SSF systems (Knight et al. 2003). Therefore, if left unattended, SSF systems have the potential for mosquito outbreaks and potential disease transmission.

Vegetation Selection

Vegetation is the key component of the wetland that provides water filtration. The purification of water can be attributed to the natural processes provided by vegetation. Each plant, for the purpose of water sanitation, will provide a benefit to the function of the wetland. This is not the case for mosquito production. Certain plant species are more conducive to mosquito breeding than others (Collins and Resch, 1989; Knight et al. 2003;

Andrews and Pollard, 2008). A list has been created by Collins and Resch which ranks the top vegetation inhabitants and assigns them a score according to mosquito production. Plants that have been found to limit mosquito breeding should be used in place of those plants found to promote mosquito breeding. The replacement of mosquito plants with non- mosquito plants will not affect water filtration or wetlands performance; these plants will perform the same as their counterparts, but with reduced concern for mosquitoes (Collins and Resch. 1989; Knight et al. 2003).

In the vegetation section of this document, the method for these values is discussed. A full listing of wetland vegetation can be found in appendix VI.

Pond Configuration- Wetland Cell Design

Proper cell design should be addressed during the construction phase of the wetland. A proper design will prove invaluable for vegetation and mosquito control. Each cell should be created using a simple design proven to aid in mosquito control. The model provided by the EPA (1998) will aid in controlling emergent vegetation, therefore reducing breeding areas for mosquitoes. In addition, wave action will be increased with open water area maximized, and predatory fish will prove effective with increased access to larval habitat.

The first priority for pond configuration is the prevention of vegetation, especially that which promotes mosquito breeding (Knight et al. 2003). Shallow water areas allow for vegetation growth and mosquito larvae development. Water depths of three feet or greater are recommended, with zones of five feet being ideal (Knight et al. 2003; Andrews and Pollard, 2008; EPA, 1998; Marin and Sonoma Mosquito Control, 2000; Collins and Resch, 1989; Mitsch and Gosselink, 1999). Water depths at three or more

feet greatly reduce emergent vegetation, allow for redistribution of shallow water near edges, enhance the oxygen content of the water through wind disturbance, and provide protective habitat for valuable predatory mosquito fish. Ponds with inadequate deep zones will not only promote mosquito breeding, but reduce the water treatment potential and hydraulic efficiency of the wetland (Knight et al. 2003).

With adequate deep zones, emergent vegetation is limited to the cell boundaries, or shore line. This will help with vegetation management, but limits wildlife habitat and cosmetic appeal. Islands of vegetation can be created within the cell to increase the vegetation to open water ratio. In areas where islands are created to help establish vegetation, some considerations should be taken to limit unwanted growth originating from the island. Islands slope should be perpendicular to the cell bottom, using a 4 ft. rise:1 ft. run ratio for vertical sides adjacent to the deep water zone. This will isolate the vegetation to the island and eliminate the chance for vegetation to spread towards the edge of the cell (EPA, 1998; 2005).

Pond boundaries should adopt the same concept in construction. The first five feet of land inundated with water in a wetland is called the pond margin. Typically this area is shallow and is where the majority of emergent vegetation exists. These margins will either inhibit or enhance mosquito breeding depending on the slope of the grade. It is recommended the first five feet adopt a 2 ft. rise:1 ft. run to 4 ft. rise:1 ft. run slope ratio for the first 5 feet or more of depth (Knight et al. 2003; Andrews and Pollard, 2008; Walton, 2003). Ponds design should limit significant shallow areas of 2.5 feet deep or less. After the pond margin, steep, almost vertical grades should be utilized adjacent to deeper water zones, similar to the island construction discussed above (Figure 8). Steep

embankments adjacent to deep water zones are ideal for mosquito control without creating concern for performance considerations. With the formation of steep embankments, initial construction cost is less and mosquito production is limited while allowing the wetland to perform as designed (Walton, 2003; Marin and Sonoma Mosquito Control, 2000; Knight et al. 2003).

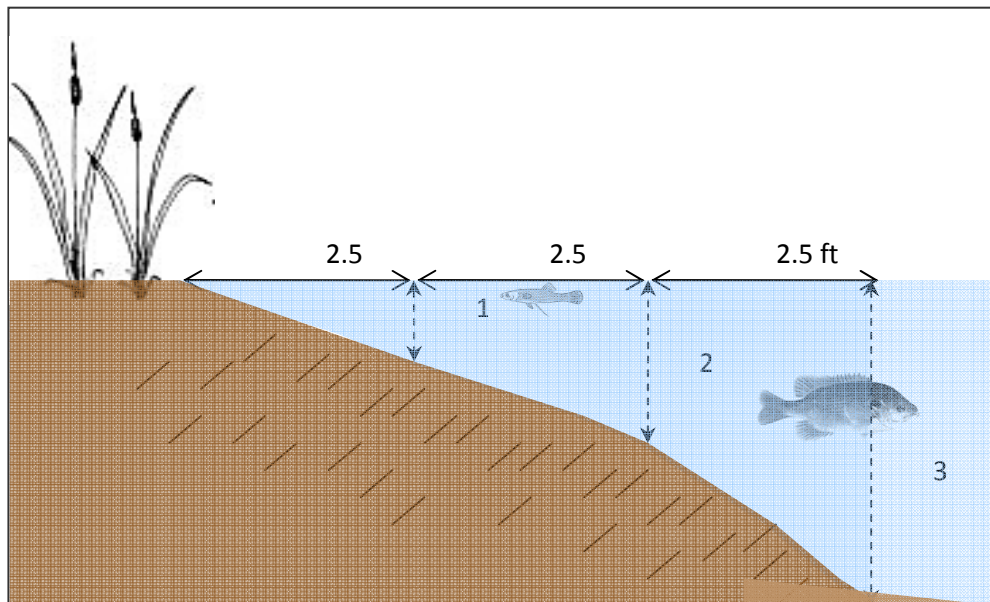


Figure 8. Side cutout of pond margin showing 2.5:1 sloping to recommended minimum depth of three feet. Photos courtesy of Clark County Wetlands Park Nature Preserve. (WPNP)

Levee construction is an important aspect of wetland cell design and should not be overlooked. They are integral in containing the water and ensuring the land is being used as intended. Permanent levees are preferred over temporary due to their ability to withstand harsh weather conditions and a cheaper maintenance cost. The following are recommendations for levee construction (Knight et al. 2003; CSU, 2008; EPA, 1998):

- Soils should consist of materials that are easily compacted. This may include clay or silt clay. Generally sandy or organic soils erode quickly and cannot be compacted. Concrete based product can be used in areas where aesthetics are not a concern. They provide durable barriers without the concern for erosion.
- Levees should be constructed at minimum of 12 feet in width. This will ensure access for management equipment, vector control vehicles, and other support resources.
- Side slopes should be constructed with a 4ft rise:1ft run ratio to deter burrowing mammals.
- The levee should be constructed at minimum one foot above planned flooding depth. This will eliminate full capacity concerns and allow flexibility within the wetlands for peak flow seasons.

In summary, a primary goal of any artificial wetland should be to reduce mosquito breeding during the design phase. Mosquitoes will occupy a large range of habitats, therefore complete eradication is impossible. Given that information, the best solution is to implement recommendations which have proven to help reduce mosquito population and aid in the abatement of mosquitoes. The following list was created by William E. Walton (2003) and provides general recommendations for enhancing mosquito abatement efforts within the constructed wetland. This information provides a follow-up to the information contained throughout the preceding section.

- Incorporate wide embankments to allow drivable shoreline access to all wetland cells. Access should have adequate turning areas. If the cell exceeds approximately 20 feet across, vehicular access should be provided on both sides.

The top of the embankment should be no less than 13 feet wide and have side slopes no steeper than a 4:1 ratio for mowing and sampling.

- Incorporate deep water zones that are free of emergent and aquatic plants. Nearly vertical edges at the perimeter of the wetland will limit growth of emergent vegetation, but may pose a safety concern.
- Provide access structures with appropriate slopes to cross deep water zones. Boats or amphibious vehicles may need to be launched in these zones for application of mosquito control agents or equipment for vegetation control.
- Keep embankments and all wetland areas free of power lines, trees, and other tall vegetation and obstructions that may limit aerial mosquito agent applications.
- Limit the width of emergent plant zones to facilitate access by predaceous fish and for application of chemical control agents.
- Compartmentalize the wetland so that the maximum width of the ponds does not exceed two times the effective distance of land based application technologies. This design feature should reduce the costs of mosquito abatement by focusing mosquito abatement on small regions of the wetland and eliminating the need to apply mosquito control agents by aircraft.
- Minimize fluctuations in water level to prevent large areas of intermittently flooded substrate or isolated pools from being created, particularly during the period of annual mosquito breeding (March to October in Clark County).
- Budget for periodic vegetation maintenance and vector control.
- Have an emergency plan that provides for immediate drainage into acceptable areas if a public health emergency occurs.

CHAPTER 5

VEGETATION

Emergent vegetation is the critical component of the water treatment process in artificial wetlands. The presence of vegetation results in cleaner water through a number of natural processes provided by the abundant plant life (CSU, 2008). The presence of organic carbon for microbial biotransformation, reduced water flow for the settling of solids, enhanced pollutant absorption, increased oxygen concentration, moderation of water temperature, and wildlife habitat can all be attributed to the vegetation within the wetlands (EPA, 2005; SWS, 2009; Knight et al. 2003). However, when vegetation becomes concentrated, mosquito larvae are protected from physical disturbance and predators. Natural mosquito deterrents, such as flowing water and wind disturbance, are eliminated which allow for the production of mosquitoes. Access for predatory fish to mosquito eggs, larvae, and pupae is diminished. Additionally, mosquito abatement efforts become more difficult and limit the effectiveness of chemical applications from lack of penetration to critical areas.

Mosquito control professionals continually stress the importance of open water and vegetation control in an effort to reduce mosquito breeding. The foremost goal of routine vegetation management is to create and maintain open water areas that are unfavorable to immature mosquito development and minimize the number of resting areas for adult mosquitoes. Emergent vegetation should be restricted to small islands that encompass no more than 50% of the open water (Andrews and Pollard, 2008). This 50:50 ratio will allow for adequate water treatment and habitat refuge for wildlife while accommodating the efforts of mosquito control. Typically, wetlands that work to minimize large stands of

emergent vegetation find that mosquito levels from the area are within an acceptable range.

Control Measures

The three most abundant plant species in a wetland setting are various Cattail species, *Typha spp.*, various Bulrush species, *Scirpus spp.*, and the common reed, *Phragmites communis* (Knight et al. 2003, Andrews and Pollard, 2008). All three of these plant species rank at the bottom of desired plants used in a wetland setting. The growth and structure of these plants promote mosquito breeding and prevent natural barriers against mosquito breeding. The ranking system created by Collins and Resh, 1989, will be discussed in greater detail later in the section.

Below are strategies for a vegetation management plan which encourage healthy, productive wetlands while effectively reducing the potential for mosquito breeding. This information has been used with permission of Andrews and Pollard (2008):

Harvesting should be considered when the source of mosquitoes is due to congestion of waterways caused by the emergent vegetation. Physical removal of the entire culpable vegetation may result in reduced congestion of waterways, change in hydrologic resistance, and allow for the planting of more desirable plant species. By eliminating emergent vegetation stands, areas where mosquitoes rest are eliminated. This will immediately make the area less hospitable for female mosquitoes. This type of management often involves the use of properly suited heavy equipment, such as amphibious backhoes or bulldozers, to physically remove vegetation (Figure 9).



Figure 9. Example of equipment needed for harvesting wetland vegetation. Image from www.ct.gov/mosquito

Due to the delicate nature of the soil within a wetland, certain effects should be accounted for. With the weight of heavy equipment, a fair amount of soil compaction is likely. This can create uneven areas in the soil where water can accumulate and support immature mosquitoes. This should be considered prior to harvesting, and plans should be made to backfill all trenches and depressions to prevent further mosquito breeding.

Pruning works to facilitate access for predatory fish, improve the flow of water, and increase open water area with the intent to limit potential mosquito breeding sites. Pruning should not be considered a long term solution. During the peak growing season, new growth will quickly replace pruned vegetation. With strategic planning during peak growing seasons, pruning will effectively reduce mosquito breeding, thereby reducing mosquito borne disease outbreaks. This planning should include areas with higher

vegetation density. For best results from pruning, both the peak growing season and peak mosquito season should be accounted for.

Edging is similar to harvesting, but the efforts are concentrated on the edges of the pond where water tends to be stagnant and shallow, both ideal conditions for mosquito oviposition. The benefits of edging include increased access for mosquito control personnel, increased wind action, and heavy equipment may not be required to remove the vegetation. Simple equipment such as weed eaters and trimmers may effectively remove the desired amount of vegetation around the edge of the pond.

If not already available, an evaluation of the potential risks and benefits of herbiciding as a vegetation management strategy may be conducted to evaluate such factors as water quality and potential toxicity to wildlife, desirable plant species, and the public. Currently, the Lake Mead Exotic Plant Management Team of the National Park Service uses an aquatically approved and EPA registered herbicide. That product may be a potential candidate for managing vegetation; however, more research is needed to make that determination. Notwithstanding, application of herbicides should be considered only as a last resort.

Burning is a method of source reduction using the destructive effects of fire. Areas of heavy vegetation are burned to ground level under close supervision of controlled burn experts. In the event this method of control is used, local fire departments, building inspectors and air quality personnel should be consulted for an expert opinion in the safety and effectiveness of the burn.

One may consider the alternative of vegetation replacement following removal of nuisance vegetation. Careful selection of plant species should be made to avoid future

problems. Various wetland plants have been ranked according to their compatibility with the goals of mosquito management (Appendix VI). Using a scoring system created by Collins and Resch (1989), 4 parameters were used to rank each plant in a wetland setting. A point system between 1 and 5 was used to score each of the four parameters, culminating in a total score for each plant. Knight, et al (2003) furthered the research by describing each of the four parameters. They are as follows:

1. Intersection line value. This value is high for plants with many stems and leaves that pass through the water surface (menisci) and lower for plants with a simple structure and few stems.
2. Crayfish food value. This value is low for plants that are preferred food for crayfish and high for plants that are not palatable or accessible to crayfish.
3. Waterfowl food value. This value is low for plants that are preferred food for waterfowl and high for plants that are not grazed by waterfowl.
4. Fish obstruction value. This parameter has a high value for plants that block fish access and low for plants with a simple structure and wider spacing that does not block fish access.

Once each plant had been given a cumulative score, they were placed into one of three categories. Categorical placement depended entirely upon the overall point total. Lower scores indicate a more suitable plant choice with regards to limiting mosquito production. The ranges of scores are as follows:

1. Scores less than 9- Lowest impact for mosquito breeding. Plants in this range are considered an optimal choice for a wetland setting.

2. Scores between 9 and 13- Acceptable levels of mosquito productivity. Low coverage of this plant is considered acceptable.
3. Scores 14 and above- Supports mosquito breeding. Presence of this plant should be minimized.

The entire list of plant species and their ranking provided by Collins and Resch can be found in Appendix VI.

With any vegetation management, removal of floating debris is a critical component. Allowing cut vegetation to accumulate on the surface will create new mosquito breeding pockets; similar to if the original vegetation was still present. Floating debris will remove wind action and cause water to stagnate creating ideal oviposition sites within the wetlands. This should be accounted for with any management plan to ensure complete source reduction in an area. The removal of the debris should be considered part of the initial plan for vegetation management rather than an afterthought.

Source Reduction

Source reduction is a term that is used interchangeably with vegetation management. Typically, the source of mosquito breeding is attributed to large stands of emergent vegetation which promote mosquito production and impedes mosquito abatement efforts. However, source reduction should also include the maintenance of pond depths, embankments, and levees to limit vegetation stands.

It is important to maintain those structures that direct water in the desired direction. Water that is allowed to escape the wetlands system will pose similar mosquito breeding concerns to that of unmaintained ponds. This water will stagnate and undesirable vegetation will emerge, creating ideal habitat for mosquitoes. Directional structures, both

natural and man-made, should be maintained to function as designed and limit nuisance water.

Pond depths should be maintained at the grade as originally constructed. This is vital the control of emergent vegetation and mosquitoes (Knight et al. 2003). With the degradation of plants and the accumulation of organic materials, the grade of a pond and overall depth will decrease, allowing vegetation to grow (Andrews and Pollard, 2008). Growth of the vegetation that promotes mosquito breeding will eventually work to fill in the pond and create an unfavorable ratio of open water to vegetation. Steep, sloping sides of the pond should be constructed and, more importantly, maintained to avoid the thick growth of plants. Dredging of cell basins may be considered to re-establish ideal pond depths and configurations if vegetation management is not successful.

CHAPTER 6

METHODOLOGY

Documents detailing the design and construction of wetlands do exist in other states and counties (Collins & Resh, 1989, Knight et al. 2003, Russell, 1998). However, Clark County, Nevada has no such document. Through the review of published articles, fact sheets, and other state and county requirements, a detailed document, specific to Clark County, can be compiled. Using well recognized resources such as the EPA, United States Army Corps of Engineers (ACOE), Society of Wetland Scientists (SWS), United States Fish and Wildlife Service (USFWS), and other leaders in various applicable fields, this document discusses wetlands construction for the control and abatement of mosquito populations.

Although the presence of mosquitoes in wetlands has been documented, validation of prominent wetland mosquito species is necessary. The confirmation of mosquitoes in the desert landscape of Clark County, Nevada will help project managers understand the importance of implementing discussed mosquito control tactics during wetland design and construction phase. Furthermore, the identification of a prominent species within the wetland is vital for the control of the overall population within the wetland. The knowledge of species- specific characteristics, including habitat, activity levels, and bloodmeal preference, allow for mosquito control professionals to plan appropriate strategies to control specific mosquitoes. Finally, the knowledge of the most common species within the wetland allows public health professionals to understand the potential disease threat posed by the wetland mosquito population. This evidence facilitates the

need for mosquito control through careful planning and execution of mosquito control practices which implement Integrated Pest Management principles.

The presence of mosquitoes in Clark County has been well documented over the last five years (2005-2009) (SNHD, 2009). Furthermore, trapping within two of the present wetland systems has occurred on a regular basis during this time. Trap counts, mosquito species, location of trapping site, and disease presence are all readily available. The data from 2009 to 2010 were used to determine that *Cx. tarsalis* and *An. Freeborni* were the top two mosquito species in the wetlands settings of Clark County, Nevada. After establishing which mosquitoes are present, the disease implication for each species was studied.

Through the combination of proven and effective guidelines for wetlands construction coupled with the validated presence of mosquitoes and disease transmission within current wetland systems in Clark County, an effective document can be created to satisfy the appeal of both mosquito abatement districts and wetland construction managers and stakeholders.

Location

Two sites were chosen to validate the presence of mosquitoes within constructed wetlands in Clark County, Nevada. Sites were chosen using the following criteria:

1. Constructed wetland
2. Available historical data
3. Purpose and/ or function
4. Location

The first site is the Wetlands Park Nature Preserve (WPNP) located at 7050 Wetlands Park Lane, Las Vegas, NV 89122. The Nature Preserve is a 3000 acre recreational wetlands situated along the Las Vegas Wash in proximity to various water treatment plants with the flow path directed towards Lake Mead. Built in 2001, The Nature Preserve features two miles of concrete walking trails, graveled secondary trails, a bird viewing blind, ponds, trail markers, and aesthetic views of the nearby alluvial fan and mountains of Rainbow Gardens. The objective of the Wetland Park project is to transform a six-mile section of the Las Vegas Wash into an interpretive desert wetland ecosystem for public use and enjoyment. In addition to creating a unique park environment, the wetlands provide for water quality improvements, slowed erosion and head cutting in the Las Vegas Wash, diversified wildlife habitat, and educational opportunities for residents and visitors (Clark County, 2010).

The WPNP is designed to serve as an recreational wetland with urban residential neighborhoods bordering the park on the west and southwest. A series of small streams interconnect five constructed ponds supplied with semi-treated effluent water, and occasionally mixed with storm and urban runoff. Various areas within the WPNP are managed to replace low-grade wildlife habitat with high-grade habitat of native vegetation. Such habitat improvements benefit a diversity of wildlife, including wetland and riparian-dependent species, and create potential habitat for a number of sensitive and endangered species (Clark County, 2010).

Historically, both staff from the University of Nevada Las Vegas (UNLV), Harry Reid Center for Environmental Studies and Southern Nevada Health District Vector Control

Program have been conducting routine disease surveillance in the area through the use of Encephalitis Vector Surveillance (EVS) traps. Historical information is available for this site which allowed for analysis of the most abundant species in the wetland and the possibility of disease transmission. This information will provide historical data to analyze the prevalence of mosquitoes within the constructed wetland setting (Andrews and Pollard, 2008).

The second site is the Henderson Bird Viewing Preserve (HBVP) located at 2400 Moser Drive, Henderson, Nevada 89011. The HBVP is part of the Kurt R. Segler Water Reclamation Facility. This facility is a 140 acre water treatment plant consisting of nine accessible ponds for bird and wildlife viewing and 13 inaccessible ponds used in the water treatment process. Constructed in 1994, the reclamation facility provides 15% of Henderson's annual water usage and can treat up to 28 million gallons per day (City of Henderson, 2010).

The HBVP was built in 1998 after 20 years of local bird watchers using the evaporating ponds which naturally attracted migratory waterfowl. Nine of the city ponds were turned into wetlands to accommodate both bird watchers and wildlife. Currently, the HBVP is home to thousands of migratory waterfowl as well as numerous resident desert birds. The ponds are surrounded by both paved and soft surfaces. The Bird Preserve is part of a natural ecosystem where natural predators help to limit the mosquito population (City of Henderson, 2010).

Prior to an organized wetland system, the area consisted of low-lying marsh lands where water was present at various times throughout the year. This presented a problem

for mosquito control as wind action was minimal and natural predators did not exist. However, through the creation of a wetland system, these problems have been eliminated.

Similar to location one, the Southern Nevada Health District Vector Control program has been conducting routine disease surveillance in the HBVP. Identical historical information is available for this site, which allowed for analysis of the most abundant species and a description of possible disease transmission concerns.

Both of these facilities are constructed wetlands which produce mosquitoes. Each is run by a separate municipality, serve a different purpose, and vary in levels of staff maintenance. The presence of mosquitoes has been confirmed at both locations. Historical trap and population data are available for both sites for the last two years. This information will be important in looking at the top three species of mosquitoes in desert wetland settings and discussing their impact on the possibility of disease transmission.

Trap Sites

At both locations, trap sites have been established prior to conducting the research. Each of the responsible surveillance groups, WPNP staff and the SNHD, trap in the same locations throughout the mosquito season. This provides authentic, consistent data which has been repeated over the past two years providing an actual, historical representation of the wetlands area.

At location one, the WPNP, all mosquito trapping is conducted by staff as part of their wetland monitoring program. Samples are then submitted to the SNHD for recording and submission for arboviral testing, which is conducted by the State of Nevada Agricultural laboratory in Sparks, NV. The WPNP staff has identified nine trapping locations (Figure

10). Trapping occurs at these locations once a month and remains consistent throughout the year.



Figure 10. Adult mosquito surveillance stations operated in the Nature Preserve during 2009. Photo property of the Wetlands Park Nature Preserve, 2009

At location two, the SNHD has established five routine trapping sites for mosquitoes within the HBVP (Figure 11). These traps are set one time a month during mosquito season. As with location one, the mosquitoes in the trap are returned to the SNHD Vector Control laboratory where they are speciated and recorded. They are then sent to the State Agriculture laboratory for arbovirus analysis.



Figure 11- Adult Mosquito Surveillance Stations Henderson Bird Viewing Preserve. Photo Courtesy of SHND, 2010.

Trapping at both sites was conducted no more than seven days apart. The relatively close proximity of each site to one another allows factors such as weather, temperature, and lunar phase impact both sites equally, creating identical trapping environments at both sites.

Global Positioning System coordinates and a standardized mapping system are available for trap location and were used for site determination. Encephalitis Vector Surveillance (EVS) (Figure 12) traps were used to trap mosquitoes, per the CDC mosquito monitoring and surveillance recommendations.



Figure 12- EVS trap . Photo property of Central Life Sciences, 2010.

The standard operating procedures (SOP) for trapping, along with the same version of EVS trap, found in figure 12, were used at both sites. Solid carbon dioxide, or dry ice, was used as the main attractant. As the dry ice sublimates, carbon dioxide gas is released. This release mimics the breath of a warm blooded animal, drawing the female mosquito into the trap. Below the dry ice container, a one watt light bulb is powered by a 6 volt battery pack. This heat source draws the mosquito in, where a fan, powered by the same 6 volt power source, creates a draft which pulls the mosquito into the catch bag, where the samples can be collected later.

EVS traps are set in the afternoon on west facing perches. This allowed the mosquitoes to be shaded from the sun prior to pick up the following morning. Captured mosquitoes were retrieved no later than 8:00am the next day, then put into a container with dry ice to ensure the integrity of the mosquito is preserved. The SOP used by both the SNHD and the WPNP can be found in Appendix 7.

Speciation of the captured mosquitoes was conducted by staff of the SNHD. Staff members dedicated to the sorting and shipping of mosquitoes would separate mosquitoes by sex and species, and count each mosquito found in the trap, ensuring an accurate representation of the mosquitoes present. The information was then logged into a database and the samples prepared for shipping.

Samples were shipped to the State of Nevada Department of Agriculture Laboratory for arboviral testing provided by West Nile Virus Grant Funding. Samples were submitted in individual species pools of 50. As a measure of checks and balances, staff members from the State of Nevada Department of Agriculture Laboratory would choose

mosquito pools at random to verify the correctness of the pool size and species contained within prior to testing.

Staff from the WPNP would sort and speciate mosquitoes captured on site under the SOP used by the SNHD. Upon receiving the samples from the WPNP staff, mosquito pools would be chosen at random to ensure correctness prior to submission for arboviral testing.

Statistical Analysis

Starting in March of 2009, archived trap logs from both locations containing trapped mosquito counts and species were compiled into an Excel spreadsheet. These results were used to determine overall mosquito counts from both sites and provide evidence for effective mosquito control through landscape design. Student's T-test was used to compare the abundance of mosquitoes between both locations, as well as the abundance of each site between the two experimental years. The difference between the two most dominant species between these two sites was determined using the two-way analysis of variance (ANOVA). The two-way ANOVA was also used to compare the abundance of all species found in these two wetlands. The T-test and ANOVA were performed using SAS® (Version 9.2, SAS Institute Inc. Cary, NC).

For this research, assumptions have been made to allow for the use of the statistical analysis. First, both sample sets were considered independent of one another due to the distance between sites. Second, both sites use reclaimed water that has undergone primary and secondary water treatment processes; therefore it was assumed the pH of the water at both sites was not a factor. Furthermore, the reclaimed water at both sites is not subjected to sanitation by chlorination. Finally, weather dependant variable were not

considered a factor due to the relative distance between sites. Each location was affected equally by any changes in weather patterns, lunar phase, or temperature.

CHAPTER 7

RESULTS

The comparison of total mosquito counts during the two year trapping period varied considerably between locations. Location one produced a considerable amount of mosquitoes (n=4561) compared to location two (n=498). Figure 13 provides visual representation of the overall difference in mosquito populations per site and the use of two tailed t-tests verified the difference was statistically significant (T-test, DF = 28, $t = -4.26$, $p = 0.0008$) between location one (39.2/site/trap) and location two (7.2/site/trap). Figure 13 shows the abundance of mosquito individuals per site per sampling event from 2009 to 2010.

Speciation of mosquito trap samples showed a diverse population with seven species of mosquito identified at both locations. Figure 14, shows a visual breakdown of the seven species and their abundance in the combined captures from both sites. It was vital to determine *Culex tarsalis* as the most common mosquito at 56% of the total mosquito count. This information was used to compare species specific production between both sites.

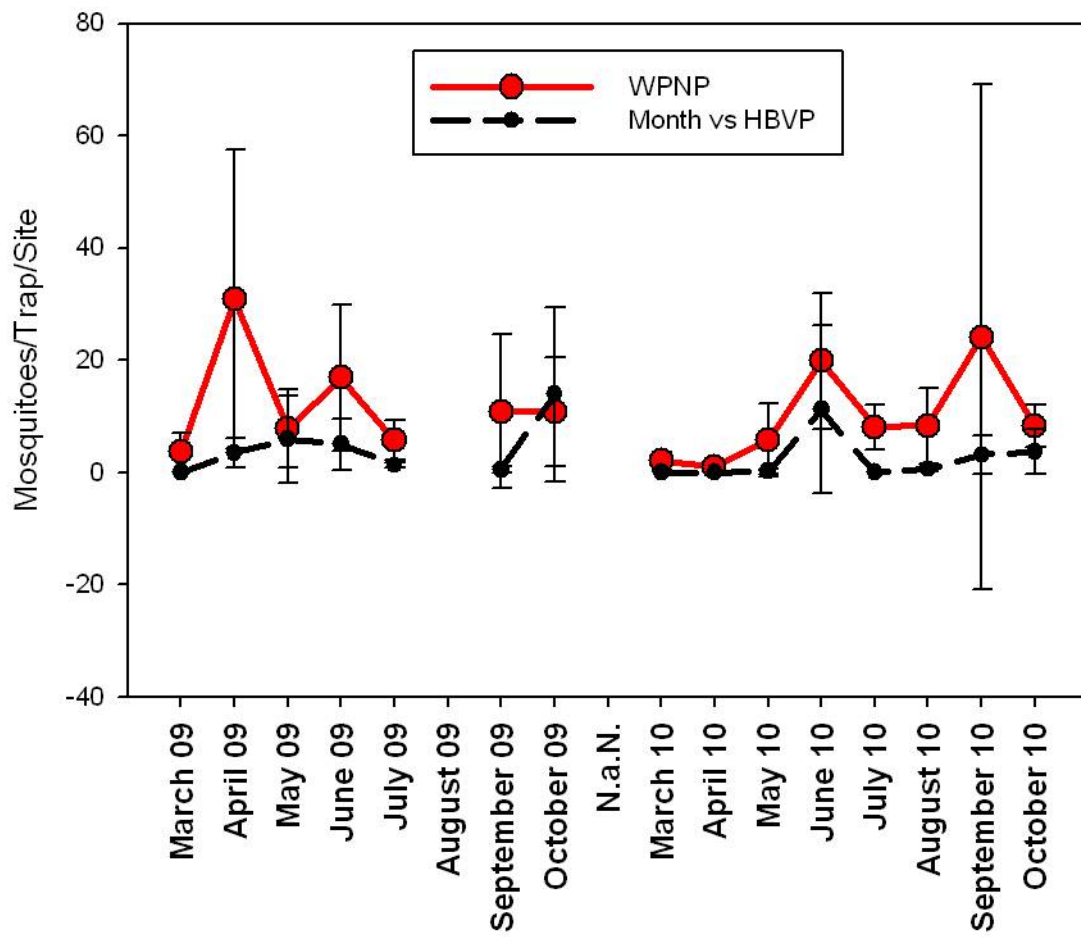


Figure 13. Abundance of mosquitoes (individuals/trap/site: Mean \pm stdev) in the two wetlands from 2009 to 2010 (N.a.N.: Not a Number); T-test, DF = 28, $t = -4.26$, $p = 0.0008$; Site 1: N= 4561, Site 2: N= 498.

Mosquito Species in % of Overall Captures Combined HBVP & WPNP 2009-2010

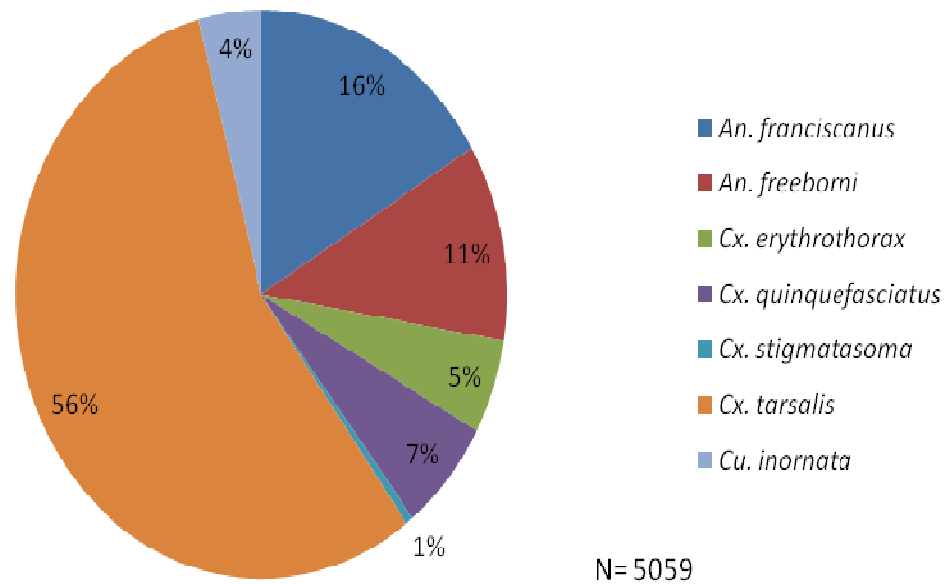


Figure 14. Mosquito species in percentage of overall capture combined from both sites, 2009-2010.

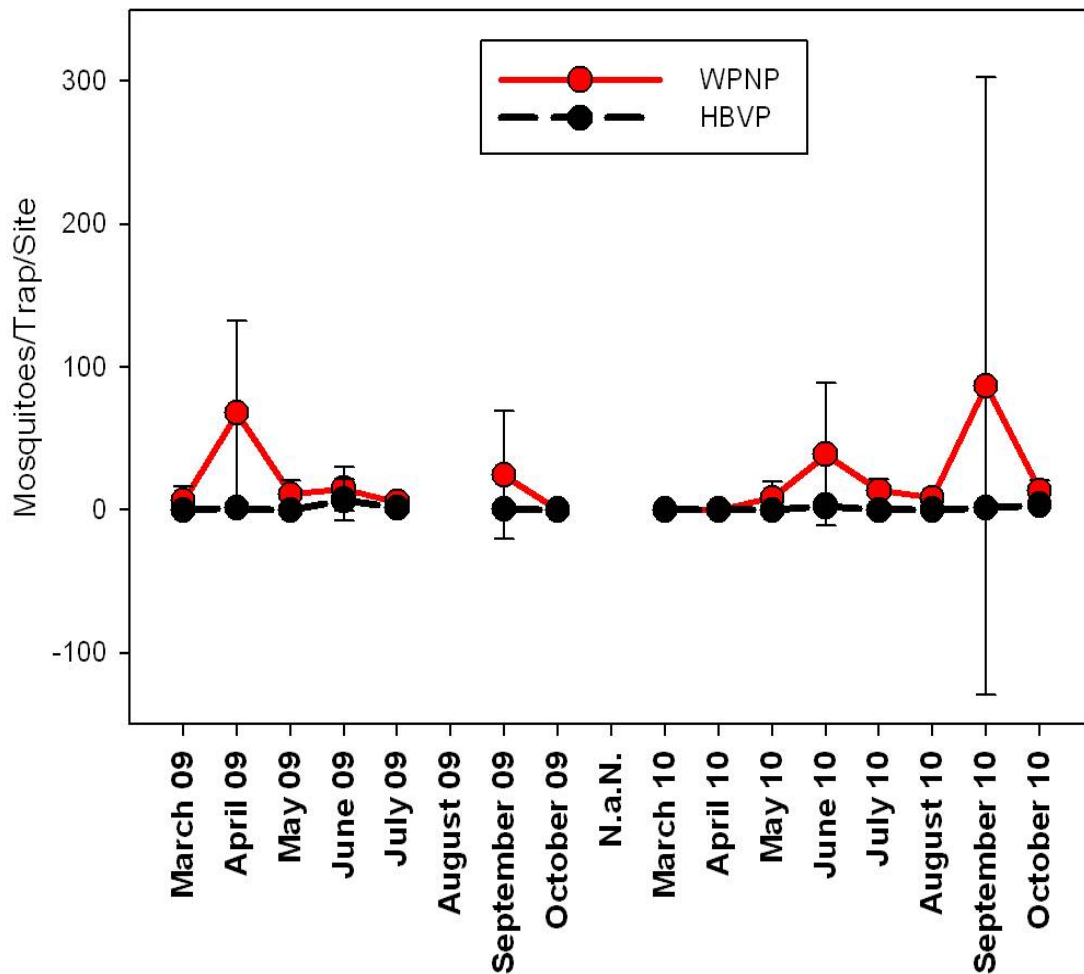


Figure 15. Abundance of *Culex tarsalis* (individuals/trap/site: Mean \pm stdev) in the two wetlands from 2009 to 2010 (N.a.N.: Not a number); T-test, DF = 28, $t = -2.78$, $P = 0.009$; Site 1 N=2741 , Site 2 N= 88

With the most abundant species determined, the Student's t-test was used to provide evidence that *Cx. tarsalis* populations would be higher from location one (20.9/trap) compared to location two (1.7/trap) (T-test, df = 28, $t = -2.78$, $P = 0.009$). Figure 15 provides visual representation of *Cx. tarsalis* abundance at each site.

In an effort to identify the most dominant species, the second most abundant species, *Anopheles freeborni* was analyzed using the Student's T-test to confirm statistical significance between sites, as with *Cx. tarsalis*. Results from the two-way ANOVA showed the population of *Cx. tarsalis* was significantly higher than the *Anopheles freeborni* population between these two wetlands [$F(2,59) = 8.7, p=0.0005$]. Post Hoc analysis using the Student-Newman, Keuls (SNK) test indicated significant difference between species overall ($df = 1, p = 0.02$). This provides confirmation that *Cx. tarsalis* is the most abundant species within both wetland locations. Two-way ANOVA and post hoc comparison also revealed that *Cx. tarsalis* was more abundant than all other species but *A. freeborni* was not significantly different from other species (SNK-test, $df=6, p < 0.0001$) and these species were more abundant in WPNP (5.7 mosquitoes/trap) than in HBVP (1.1 mosquitoes/trap) (SNK-test, $df = 1, p = 0.0002$).

A summary of overall abundance for each mosquito species per trap- per site can be found in table one.

Table 1. Summary of abundance of each mosquito species (per trap per site).

Location	Year	Species	Minimum	Maximum
WPNP	2009	<i>Anopheles franciscanus</i>	2.3	25.8
WPNP	2009	<i>Anopheles freeborni</i>	1.1	10.5
WPNP	2009	<i>Culex erythrothorax</i>	0.0	1.0
WPNP	2009	<i>Culex quinquefasciatus</i>	1.0	18.3
WPNP	2009	<i>Culex tarsalis</i>	2.0	76.5
WPNP	2009	<i>Culiseta inornata</i>	1.0	13.1
WPNP	2010	<i>Anopheles franciscanus</i>	0.3	20.4
WPNP	2010	<i>Anopheles freeborni</i>	0.6	19.7
WPNP	2010	<i>Culex erythrothorax</i>	1.0	9.0
WPNP	2010	<i>Culex quinquefasciatus</i>	1.0	6.0
WPNP	2010	<i>Culex stigmatasoma</i>	0.0	1.0
WPNP	2010	<i>Culex tarsalis</i>	1.0	86.8
WPNP	2010	<i>Culiseta inornata</i>	1.0	3.5
HBVP	2009	<i>Anopheles franciscanus</i>	0.0	3.2
HBVP	2009	<i>Anopheles freeborni</i>	0.0	0.3
HBVP	2009	<i>Culex quinquefasciatus</i>	1.0	4.5
HBVP	2009	<i>Culex stigmatasoma</i>	1.0	8.0
HBVP	2009	<i>Culex tarsalis</i>	0.5	11.7
HBVP	2009	<i>Culiseta inornata</i>	0.0	14.5
HBVP	2010	<i>Anopheles franciscanus</i>	0.2	0.4
HBVP	2010	<i>Culex erythrothorax</i>	0.6	30.5
HBVP	2010	<i>Culex quinquefasciatus</i>	1.0	9.0
HBVP	2010	<i>Culex tarsalis</i>	1.0	3.6
HBVP	2010	<i>Culiseta inornata</i>	0.0	2.0

CHAPTER 8

DISCUSSION

This study assessed mosquito abundance and diversity between two wetland systems with different designs. It was determined that wetlands within Clark County, Nevada engineered according to EPA guidelines, does limit vegetation and enhance natural predation of mosquito larvae, thereby reducing the diverse mosquito populations within the local wetland systems, as evidenced from the significantly lower abundance of mosquitoes in the HBVP.

The hypothesis that location one would produce more mosquitoes than location two due to differences in design was supported by the findings. When comparing overall mosquito populations, location one produced a significant amount of mosquitoes when compared to location 2 (Figure 13). This data provides evidence that a properly designed wetland can reduce the overall mosquito populations within a wetland, thereby reducing the efforts of the organized or assigned mosquito control & monitoring program.

These results parallel the expectations of the given landscapes for each location. The WPNP contains more slow moving, congested waterways caused by overgrown flora. Natural processes which increase soil accumulation have altered the desired pond depths and slopes, enabling plant life to encroach on the pond, minimizing open surface area. In turn, this reduction in surface area has limited natural control, such as wave action and predation, allowing for uncontested larval development. *Gambusia affinis*, or mosquito fish, populations are present within the wetland system; however they are not as prolific as in the past. Other factors including flooding, water tables, and recent construction also increase mosquito numbers throughout the park.

Location 2, the HBVP, was constructed and is managed under the guidelines set by the EPA. Each pond is deep with maintained shoreline vegetation. Surface area on each pond is maximized, allowing for natural larval reduction through large populations of *Gambusia affinis* and other predators. Cattail stands are present in the HBVP, but are isolated to one pond and maintained for avian habitat. These stands are kept in the middle of the pond and have completely vertical sides. This feature, as recommended by the EPA, allows the size of the stand to be controlled, limiting habitat for larval development.

Both sites are labeled and function as wetlands. The WPNP is a recreational area with a traditional wetland landscape. The park is managed to maintain the aesthetics of the park in addition to managing mosquito populations. The HBVP is a water treatment plant that doubles as a bird viewing preserve. A visual assessment of each property would lead one to believe the WPNP would produce more mosquitoes based on the landscape alone. However, during the peak summer months, both locations produce mosquitoes as shown by the trap counts from the sampling period.

Seventeen species of mosquitoes are local to Clark County (SNHD, 2009). Six of the seventeen species are considered rare and seldom found during routine surveillance efforts. Four species are geographically isolated to restricted habitats, none of which are wetlands. Analysis of mosquito populations from both wetland systems identified the remaining seven species of mosquitoes at both locations: *Anopheles franciscanus*, *Anopheles freeborni*, *Culex erythrothorax*, *Culex quinquefasciatus*, *Culex stigmatasoma*, *Culex tarsalis*, and *Culiseta inornata*. Forty one percent (7 of 17) of the resident mosquitoes in Clark County reside within the wetlands habitat. The presence of seven

species in the wetland supports the second hypothesis that local wetlands maintain a species rich mosquito population.

Further analysis of mosquito populations concluded that *Culex tarsalis* is the dominant mosquito species among wetland locations in Clark County, Nevada. Population numbers from the WPNP show the *Cx. tarsalis* population is significantly higher than the *An. freeborni* population. Results showed *Cx. tarsalis* counts made up a significant amount of all mosquitoes captured. Furthermore, *Cx. tarsalis* populations from the WPNP outnumbered populations from the HBVP 20:1. These data reinforce the concept that proper design limits mosquito populations while also reducing the potential for disease transmission. *Cx. tarsalis* is the single most important mosquito vector in Clark County and, according to this study, the most abundant in local wetland environments.

Cx. tarsalis is considered the most important vector of arboviruses in western North America, responsible for maintenance, amplification and epidemic transmission of West Nile Virus (WNV), St. Louis Encephalitis (SLE), and Western Equine Encephalitis (WEE) (Rutgers University, 1993). In 2005, local *Cx. tarsalis* pools tested positive for WNV, SLE, and WEE (SNHD, 2006). During the past 5 years, both experimental sites have produced WNV and/ or encephalitis positive *Cx. tarsalis* mosquitoes. The presence of disease and capable mosquito vectors is the primary reason EPA guidelines should be instituted for wetland construction to reduce mosquito populations.

The biting habits of *Cx. tarsalis* initiate the cause for concern. In spring, when population abundance is low, most females feed primarily on Passeriformes birds. Many of these birds are potential hosts for WNV or other encephalitis viruses, enabling the female mosquito to become a vector. During late summer when mosquito

populations are high, bird avoidance behavior diverts many female mosquitoes to feed on humans as well as other mammals including horses, cattle, and rabbits. This host shift is a key element in virus transmission among *Culex* mosquitoes and man.

Cx. tarsalis can be found in almost every environment west of the Mississippi (Rutgers University, 1993). With the ability to colonize standing freshwater, *Cx. tarsalis* can quickly become the majority of any mosquito population. This species thrives in water where micro-floral blooms are produced by the release of nutrients from decomposing vegetation, a cycle very common in wetlands with established shoreline vegetation. With a number of factors, including lack of natural limitation and ideal habitat, the control of *Cx. tarsalis*, and all mosquitoes, within the wetland can be difficult and expensive post construction.

The *Culex* species are considered the main vector for many arboviruses worldwide (CDC, 2003). However, all mosquito species found in the wetland are a potential threat for disease transmission, especially WNV (AMCA, 2009). The second most abundant species, *An. freeborni*, is known as the Western Malaria mosquito for its ability to transmit and host malarial parasites. Visitors from all over the world, including Malaria endemic areas, vacation in Las Vegas, NV. The combination of competent vectors and potential hosts increases the chance for malarial transmission in Clark County. In the event a case of malaria was confirmed in the area, the effect on an already struggling economy could be devastating. This concept is unlikely, but in theory, the potential exists.

In addition to disease transmission, all mosquitoes have a nuisance factor. This affects residents near the wetland, as well as visitors and workers within the wetland. Dispersal

is primarily during host-seeking flights, which can range up to 17 miles over the life of a female mosquito (Rutgers, 1993). With each species capable of seeking a bloodmeal well beyond the constraints of the breeding site, it is important to limit mosquito breeding during larval development. Larval control is considered the best and most effective means of controlling adult mosquito populations due to concentration and habitat identification. Once a mosquito emerges, it is much more difficult to control and eradicate because of dispersal capabilities.

A brief review of the data provide evidence of how effective a properly constructed wetland can be at reducing overall mosquito numbers, and more so, reducing the number of primary disease vectors present in Clark County, Nevada. This reduction in mosquitoes can be attributed to the control within wetland sites of Clark County, Nevada. Vegetation is beneficial; however overgrown, thick stands of water reduce flow and create stagnant bodies of water which limit the wetland and its ability to function. For a wetland to succeed, both in the reduction of mosquito populations and overall efficiency, the control of vegetation should be the number one priority. This goal is achievable through the implementation of the designs discussed in this paper.

Through the implementation of EPA guidelines, similar results have been obtained in other landscapes, in other states. It should be expected that EPA suggested practices will provide comparable results, regardless of location.

Final Thoughts

Mosquito species have evolved to exploit a wide variety of habitats. Because mosquitoes are a natural part of wetland ecosystems, permanent and total elimination of mosquitoes from wetlands is not a realistic goal. However, current scientific

understanding supports the position that environmentally-compatible measures can be taken to minimize mosquito production from artificial wetlands (SWS, 2009). Mosquito control begins with the use of Integrated Pest Management concepts that encourage ecological diversity and natural mosquito predators, while minimizing the creation of site features which promote mosquito production.

Wetlands are fragile systems which provide crucial environmental processes and socio-economic functions (SWS, 2009). As society is confronted with new and emerging mosquito-borne diseases, the need to protect human health and wetland function will only increase. If the sustainment of wetlands through a “no net loss” initiative is going to be successful, then wetland professionals must address the need for mosquito control through design standards in pre- construction planning.

Wetland professionals, regulatory agencies, public health organizations, and mosquito control agencies should consult with one another and the public during the planning, design, implementation, management and maintenance phases of wetland creation, restoration or enhancement projects. Mosquito control efforts can be greatly minimized if the wetland is constructed in a manner to reduce breeding habitat, or is conducive to modern control methods by allowing access. All wetland projects must include the minimization of mosquito production within the scope of their design, regardless of how it is achieved. This document outlines and provides evidence of how pre construction planning and the implementation of proven design standards will aid in limiting mosquito populations within the wetland system.

APPENDIX I

SECTION 404 OF THE CLEAN WATER ACT

Title 33 - navigation and navigable waterschapter 26 - water pollution prevention and controls
subchapter iv - permits and licenses

(a) Discharge into navigable waters at specified disposal sites

The Secretary may issue permits, after notice and opportunity for public hearings for the discharge of dredged or fill material into the navigable waters at specified disposal sites. Not later than the fifteenth day after the date an applicant submits all the information required to complete an application for a permit under this subsection, the Secretary shall publish the notice required by this subsection.

(b) Specification for disposal sites

Subject to subsection (c) of this section, each such disposal site shall be specified for each such permit by the Secretary (1) through the application of guidelines developed by the Administrator, in conjunction with the Secretary, which guidelines shall be based upon criteria comparable to the criteria applicable to the territorial seas, the contiguous zone, and the ocean under section 1343(c) of this title, and (2) in any case where such guidelines under clause (1) alone would prohibit the specification of a site, through the application additionally of the economic impact of the site on navigation and anchorage.

(c) Denial or restriction of use of defined areas as disposal sites

The Administrator is authorized to prohibit the specification (including the withdrawal of specification) of any defined area as a disposal site, and he is authorized to deny or restrict the use of any defined area for specification (including the withdrawal of specification) as a disposal site, whenever he determines, after notice and opportunity for public hearings, that the discharge of such materials into such area will have an unacceptable adverse effect on municipal water supplies, shellfish beds and fishery areas (including spawning and breeding areas), wildlife, or recreational areas. Before making such determination, the Administrator shall consult with the Secretary. The Administrator shall set forth in writing and make public his findings and his reasons for making any determination under this subsection.

(d) "Secretary" defined

The term "Secretary" as used in this section means the Secretary of the Army, acting through the Chief of Engineers.

(e) General permits on State, regional, or nationwide basis

(1) In carrying out his functions relating to the discharge of dredged or fill material under this section, the Secretary may, after notice and opportunity for public hearing, issue general permits on a State, regional, or nationwide basis for any category of activities involving discharges of dredged or fill material if the Secretary determines that the activities in such category are similar in nature, will cause only minimal adverse environmental effects when performed separately, and will have only minimal cumulative adverse effect on the environment. Any general permit issued under this subsection shall

(A) be based on the guidelines described in subsection (b)(1) of this section, and
(B) set forth the requirements and standards which shall apply to any activity authorized by such general permit.

(2) No general permit issued under this subsection shall be for a period of more than five years after the date of its issuance and such general permit may be revoked or modified by the Secretary if, after opportunity for public hearing, the Secretary determines that the activities authorized by such general permit have an adverse impact on the environment or such activities are more appropriately authorized by individual permits.

(f) Non-prohibited discharge of dredged or fill material

(1) Except as provided in paragraph (2) of this subsection, the discharge of dredged or fill material

(A) from normal farming, silviculture, and ranching activities such as plowing, seeding, cultivating, minor drainage, harvesting for the production of food, fiber, and forest products, or upland soil and water conservation practices;

(B) for the purpose of maintenance, including emergency reconstruction of recently damaged parts, of currently serviceable structures such as dikes, dams, levees, groins, riprap, breakwaters, causeways, and bridge abutments or approaches, and transportation structures;

(C) for the purpose of construction or maintenance of farm or stock ponds or irrigation ditches, or the maintenance of drainage ditches;

(D) for the purpose of construction of temporary sedimentation basins on a construction site which does not include placement of fill material into the navigable waters;

(E) for the purpose of construction or maintenance of farm roads or forest roads, or temporary roads for moving mining equipment, where such roads are constructed and maintained, in accordance with best management practices, to assure that flow and circulation patterns and chemical and biological characteristics of the navigable waters are not impaired, that the reach of the navigable waters is not reduced, and that any adverse effect on the aquatic environment will be otherwise minimized;

(F) resulting from any activity with respect to which a State has an approved program under section 1288(b)(4) of this title which meets the requirements of subparagraphs (B) and (C) of such section, is not prohibited by or otherwise subject to regulation under this section or section 1311(a) or 1342 of this title (except for effluent standards or prohibitions under section 1317 of this title).

(2) Any discharge of dredged or fill material into the navigable waters incidental to any activity having as its purpose bringing an area of the navigable waters into a use to which it was not previously subject, where the flow or circulation of navigable waters may be impaired or the reach of such waters be reduced, shall be required to have a permit under this section.

(g) State administration

(1) The Governor of any State desiring to administer its own individual and general permit program for the discharge of dredged or fill material into the navigable waters (other than those waters which are presently used, or are susceptible to use in their natural condition or by reasonable improvement as a means to transport interstate or foreign commerce shoreward to their ordinary high water mark, including all waters which are subject to the ebb and flow of the tide shoreward to their mean high water mark, or mean

higher high water mark on the west coast, including wetlands adjacent thereto) within its jurisdiction may submit to the Administrator a full and complete description of the program it proposes to establish and administer under State law or under an interstate compact. In addition, such State shall submit a statement from the attorney general (or the attorney for those State agencies which have independent legal counsel), or from the chief legal officer in the case of an interstate agency, that the laws of such State, or the interstate compact, as the case may be, provide adequate authority to carry out the described program.

(2) Not later than the tenth day after the date of the receipt of the program and statement submitted by any State under paragraph (1) of this subsection, the Administrator shall provide copies of such program and statement to the Secretary and the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service.

(3) Not later than the ninetieth day after the date of the receipt by the Administrator of the program and statement submitted by any State, under paragraph (1) of this subsection, the Secretary and the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service, shall submit any comments with respect to such program and statement to the Administrator in writing.

(h) Determination of State's authority to issue permits under State program; approval; notification; transfers to State program

(1) Not later than the one-hundred-twentieth day after the date of the receipt by the Administrator of a program and statement submitted by any State under paragraph (1) of this subsection, the Administrator shall determine, taking into account any comments submitted by the Secretary and the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service, pursuant to subsection (g) of this section, whether such State has the following authority with respect to the issuance of permits pursuant to such program:

- (A) To issue permits which -
- (i) apply, and assure compliance with, any applicable requirements of this section, including, but not limited to, the guidelines established under subsection (b)(1) of this section, and sections 1317 and 1343 of this title;
 - (ii) are for fixed terms not exceeding five years; and
 - (iii) can be terminated or modified for cause including, but not limited to, the following:
 - (I) violation of any condition of the permit;
 - (II) obtaining a permit by misrepresentation, or failure to disclose fully all relevant facts;
 - (III) change in any condition that requires either a temporary or permanent reduction or elimination of the permitted discharge.
- (B) To issue permits which apply, and assure compliance with, all applicable requirements of section 1318 of this title, or to inspect, monitor, enter, and require reports to at least the same extent as required in section 1318 of this title.
- (C) To assure that the public, and any other State the waters of which may be affected, receive notice of each application for a permit and to provide an opportunity for public hearing before a ruling on each such application.
- (D) To assure that the Administrator receives notice of each application (including a copy thereof) for a permit.
- (E) To assure that any State (other than the permitting State), whose waters may be

affected by the issuance of a permit may submit written recommendations to the permitting State (and the Administrator) with respect to any permit application and, if any part of such written recommendations are not accepted by the permitting State, that the permitting State will notify such affected State (and the Administrator) in writing of its failure to so accept such recommendations together with its reasons for so doing.

(F) To assure that no permit will be issued if, in the judgment of the Secretary, after consultation with the Secretary of the department in which the Coast Guard is operating, anchorage and navigation of any of the navigable waters would be substantially impaired thereby.

(G) To abate violations of the permit or the permit program, including civil and criminal penalties and other ways and means of enforcement.

(H) To assure continued coordination with Federal and Federal-State water-related planning and review processes.

(2) If, with respect to a State program submitted under subsection (g)(1) of this section, the Administrator determines that such State -

(A) has the authority set forth in paragraph (1) of this subsection, the Administrator shall approve the program and so notify (i) such State and (ii) the Secretary, who upon subsequent notification from such State that it is administering such program, shall suspend the issuance of permits under subsections (a) and (e) of this section for activities with respect to which a permit may be issued pursuant to such State program; or

(B) does not have the authority set forth in paragraph (1) of this subsection, the Administrator shall so notify such State, which notification shall also describe the revisions or modifications necessary so that such State may resubmit such program for a determination by the Administrator under this subsection.

(3) If the Administrator fails to make a determination with respect to any program submitted by a State under subsection (g)(1) of this section within one-hundred-twenty days after the date of the receipt of such program, such program shall be deemed approved pursuant to paragraph (2)(A) of this subsection and the Administrator shall so notify such State and the Secretary who, upon subsequent notification from such State that it is administering such program, shall suspend the issuance of permits under subsection (a) and (e) of this section for activities with respect to which a permit may be issued by such State.

(4) After the Secretary receives notification from the Administrator under paragraph (2) or (3) of this subsection that a State permit program has been approved, the Secretary shall transfer any applications for permits pending before the Secretary for activities with respect to which a permit may be issued pursuant to such State program to such State for appropriate action.

(5) Upon notification from a State with a permit program approved under this subsection that such State intends to administer and enforce the terms and conditions of a general permit issued by the Secretary under subsection (e) of this section with respect to activities in such State to which such general permit applies, the Secretary shall suspend the administration and enforcement of such general permit with respect to such activities.

(i) Withdrawal of approval

Whenever the Administrator determines after public hearing that a State is not administering a program approved under subsection (h)(2)(A) of this section, in

accordance with this section, including, but not limited to, the guidelines established under subsection (b)(1) of this section, the Administrator shall so notify the State, and, if appropriate corrective action is not taken within a reasonable time, not to exceed ninety days after the date of the receipt of such notification, the Administrator shall (1) withdraw approval of such program until the Administrator determines such corrective action has been taken, and (2) notify the Secretary that the Secretary shall resume the program for the issuance of permits under subsections (a) and (e) of this section for activities with respect to which the State was issuing permits and that such authority of the Secretary shall continue in effect until such time as the Administrator makes the determination described in clause (1) of this subsection and such State again has an approved program.

(j) Copies of applications for State permits and proposed general permits to be transmitted to Administrator

Each State which is administering a permit program pursuant to this section shall transmit to the Administrator (1) a copy of each permit application received by such State and provide notice to the Administrator of every action related to the consideration of such permit application, including each permit proposed to be issued by such State, and (2) a copy of each proposed general permit which such State intends to issue. Not later than the tenth day after the date of the receipt of such permit application or such proposed general permit, the Administrator shall provide copies of such permit application or such proposed general permit to the Secretary and the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service. If the Administrator intends to provide written comments to such State with respect to such permit application or such proposed general permit, he shall so notify such State not later than the thirtieth day after the date of the receipt of such application or such proposed general permit and provide such written comments to such State, after consideration of any comments made in writing with respect to such application or such proposed general permit by the Secretary and the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service, not later than the ninetieth day after the date of such receipt. If such State is so notified by the Administrator, it shall not issue the proposed permit until after the receipt of such comments from the Administrator, or after such ninetieth day, whichever first occurs. Such State shall not issue such proposed permit after such ninetieth day if it has received such written comments in which the Administrator objects (A) to the issuance of such proposed permit and such proposed permit is one that has been submitted to the Administrator pursuant to subsection (h)(1)(E) of this section, or (B) to the issuance of such proposed permit as being outside the requirements of this section, including, but not limited to, the guidelines developed under subsection (b)(1) of this section unless it modifies such proposed permit in accordance with such comments. Whenever the Administrator objects to the issuance of a permit under the preceding sentence such written objection shall contain a statement of the reasons for such objection and the conditions which such permit would include if it were issued by the Administrator. In any case where the Administrator objects to the issuance of a permit, on request of the State, a public hearing shall be held by the Administrator on such objection. If the State does not resubmit such permit revised to meet such objection

within 30 days after completion of the hearing or, if no hearing is requested within 90 days after the date of such objection, the Secretary may issue the permit pursuant to subsection (a) or (e) of this section, as the case may be, for such source in accordance with the guidelines and requirements of this chapter.

(k) Waiver

In accordance with guidelines promulgated pursuant to subsection (i)(2) of section 1314 of this title, the Administrator is authorized to waive the requirements of subsection (j) of this section at the time of the approval of a program pursuant to subsection (h)(2)(A) of this section for any category (including any class, type, or size within such category) of discharge within the State submitting such program.

(l) Categories of discharges not subject to requirements

The Administrator shall promulgate regulations establishing categories of discharges which he determines shall not be subject to the requirements of subsection (j) of this section in any State with a program approved pursuant to subsection (h)(2)(A) of this section. The Administrator may distinguish among classes, types, and sizes within any category of discharges.

(m) Comments on permit applications or proposed general permits by Secretary of the Interior acting through Director of United States Fish and Wildlife Service

Not later than the ninetieth day after the date on which the Secretary notifies the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service that

(1) an application for a permit under subsection (a) of this section has been received by the Secretary, or

(2) the Secretary proposes to issue a general permit under subsection (e) of this section, the Secretary of the Interior, acting through the Director of the United States Fish and Wildlife Service, shall submit any comments with respect to such application or such proposed general permit in writing to the Secretary.

(n) Enforcement authority not limited

Nothing in this section shall be construed to limit the authority of the Administrator to take action pursuant to section 1319 of this title.

(o) Public availability of permits and permit applications

A copy of each permit application and each permit issued under this section shall be available to the public. Such permit application or portion thereof, shall further be available on request for the purpose of reproduction.

(p) Compliance

Compliance with a permit issued pursuant to this section, including any activity carried out pursuant to a general permit issued under this section, shall be deemed compliance, for purposes of sections 1319 and 1365 of this title, with sections 1311, 1317, and 1343 of this title.

(q) Minimization of duplication, needless paperwork, and delays in issuance; agreements

Not later than the one-hundred-eightieth day after December 27, 1977, the Secretary shall enter into agreements with the Administrator, the Secretaries of the Departments of Agriculture, Commerce, Interior, and Transportation, and the heads of other appropriate Federal agencies to minimize, to the maximum extent practicable, duplication, needless

paperwork, and delays in the issuance of permits under this section. Such agreements shall be developed to assure that, to the maximum extent practicable, a decision with respect to an application for a permit under subsection (a) of this section will be made not later than the ninetieth day after the date the notice for such application is published under subsection (a) of this section.

(r) Federal projects specifically authorized by Congress

The discharge of dredged or fill material as part of the construction of a Federal project specifically authorized by Congress, whether prior to or on or after December 27, 1977, is not prohibited by or otherwise subject to regulation under this section, or a State program approved under this section, or section 1311(a) or 1342 of this title (except for effluent standards or prohibitions under section 1317 of this title), if information on the effects of such discharge, including consideration of the guidelines developed under subsection (b)(1) of this section, is included in an environmental impact statement for such project pursuant to the National Environmental Policy Act of 1969 (42 U.S.C. 4321 et seq.) and such environmental impact statement has been submitted to Congress before the actual discharge of dredged or fill material in connection with the construction of such project and prior to either authorization of such project or an appropriation of funds for such construction.

(s) Violation of permits

(1) Whenever on the basis of any information available to him the Secretary finds that any person is in violation of any condition or limitation set forth in a permit issued by the Secretary under this section, the Secretary shall issue an order requiring such person to comply with such condition or limitation, or the Secretary shall bring a civil action in accordance with paragraph (3) of this subsection.

(2) A copy of any order issued under this subsection shall be sent immediately by the Secretary to the State in which the violation occurs and other affected States. Any order issued under this subsection shall be by personal service and shall state with reasonable specificity the nature of the violation, specify a time for compliance, not to exceed thirty days, which the Secretary determines is reasonable, taking into account the seriousness of the violation and any good faith efforts to comply with applicable requirements. In any case in which an order under this subsection is issued to a corporation, a copy of such order shall be served on any appropriate corporate officers.

(3) The Secretary is authorized to commence a civil action for appropriate relief, including a permanent or temporary injunction for any violation for which he is authorized to issue a compliance order under paragraph (1) of this subsection. Any action under this paragraph may be brought in the district court of the United States for the district in which the defendant is located or resides or is doing business, and such court shall have jurisdiction to restrain such violation and to require compliance. Notice of the commencement of such action (Note: Probably should be action) shall be given immediately to the appropriate State.

(4) Any person who violates any condition or limitation in a permit issued by the Secretary under this section, and any person who violates any order issued by the Secretary under paragraph (1) of this subsection, shall be subject to a civil penalty not to exceed \$25,000 per day for each violation. In determining the amount of a civil penalty the court shall consider the seriousness of the violation or violations, the economic

benefit (if any) resulting from the violation, any history of such violations, any good-faith efforts to comply with the applicable requirements, the economic impact of the penalty on the violator, and such other matters as justice may require.

(t) Navigable waters within State jurisdiction

Nothing in this section shall preclude or deny the right of any State or interstate agency to control the discharge of dredged or fill material in any portion of the navigable waters within the jurisdiction of such State, including any activity of any Federal agency, and each such agency shall comply with such State or interstate requirements both substantive and procedural to control the discharge of dredged or fill material to the same extent that any person is subject to such requirements. This section shall not be construed as affecting or impairing the authority of the Secretary to maintain navigation.

SOURCE

(June 30, 1948, ch. 758, title IV, Sec. 404, as added Oct. 18, 1972, Pub. L. 92-500, Sec. 2, 86 Stat. 884; amended Dec. 27, 1977, Pub. L. 95-217, Sec. 67(a), (b), 91 Stat. 1600; Feb. 4, 1987, Pub. L. 100-4, title III, Sec. 313(d), 101 Stat. 45.)

REFERENCES IN TEXT

The National Environmental Policy Act of 1969, referred to in subsec. (r), is Pub. L. 91-190, Jan. 1, 1970, 83 Stat. 852, as amended, which is classified generally to chapter 55 (Sec. 4321 et seq.) of Title 42, The Public Health and Welfare. For complete classification of this Act to the Code, see Short Title note set out under section 4321 of Title 42 and Tables.

TRANSFER OF FUNCTIONS

Enforcement functions of Administrator or other official of the Environmental Protection Agency and of Secretary or other official in Department of Interior relating to review of the Corps of Engineers' dredged and fill material permits and such functions of Secretary of the Army, Chief of Engineers, or other official in Corps of Engineers of the United States Army relating to compliance with dredged and fill material permits issued under this section with respect to pre-construction, construction, and initial operation of transportation system for Canadian and Alaskan natural gas were transferred to the Federal Inspector, Office of Federal Inspector for the Alaska Natural Gas Transportation System, until the first anniversary of the date of initial operation of the Alaska Natural Gas Transportation System, see Reorg. Plan No. 1 of 1979, Sec. 102(a), (b), (e), 203(a), 44 F.R. 33663, 33666, 93 Stat. 1373, 1376,

APPENDIX II

MOSQUITO SPECIES FOUND TO BE INFECTED WITH WEST NILE VIRUS

- | | |
|---|--|
| 1. <i>Aedes aegypti</i> | 31. <i>Deinocerites cancer</i> |
| 2. <i>Aedes albopictus</i> | 32. <i>Mansonia titillans</i> |
| 3. <i>Aedes cinereus</i> | 33. <i>Ochlerotatus atlanticus/tormentor</i> |
| 4. <i>Aedes vexans</i> | 34. <i>Ochlerotatus atropalpus</i> |
| 5. <i>Anopheles atropos</i> | 35. <i>Ochlerotatus canadensis</i> |
| 6. <i>Anopheles barberi</i> | 36. <i>Ochlerotatus cantator</i> |
| 7. <i>Anopheles crucians/bradleyi</i> | 37. <i>Ochlerotatus condolezens</i> |
| 8. <i>Anopheles franciscanus</i>* | 38. <i>Ochlerotatus dorsalis</i> |
| 9. <i>Anopheles freeborni</i>* | 39. <i>Ochlerotatus dupreei</i> |
| 10. <i>Anopheles hermsi</i> | 40. <i>Ochlerotatus fitchii</i> |
| 11. <i>Anopheles punctipennis</i> | 41. <i>Ochlerotatus fulvus pallens</i> |
| 12. <i>Anopheles quadrimaculatus</i> | 42. <i>Ochlerotatus grossbecki</i> |
| 13. <i>Anopheles walkeri</i> | 43. <i>Ochlerotatus infirmatus</i> |
| 14. <i>Coquillettidia perturbans</i> | 44. <i>Ochlerotatus japonicus</i> |
| 15. <i>Culex coronator</i> | 45. <i>Ochlerotatus melanimon</i> |
| 16. <i>Culex erraticus</i> | 46. <i>Ochlerotatus nigromaculis</i> |
| 17. <i>Culex erythrothorax</i>* | 47. <i>Ochlerotatus provocans</i> |
| 18. <i>Culex nigripalpus</i> | 48. <i>Ochlerotatus sollicitans</i> |
| 19. <i>Culex pipiens</i> | 49. <i>Ochlerotatus squamiger</i> |
| 20. <i>Culex quinquefasciatus</i>* | 50. <i>Ochlerotatus sticticus</i> |
| 21. <i>Culex restuans</i> | 51. <i>Ochlerotatus stimulans</i> |
| 22. <i>Culex salinarius</i> | 52. <i>Ochlerotatus taeniorhynchus</i> |
| 23. <i>Culex stigmatasoma</i>* | 53. <i>Ochlerotatus triseriatus</i> |
| 24. <i>Culex tarsalis</i>* | 54. <i>Ochlerotatus trivittatus</i> |
| 25. <i>Culex territans</i> | 55. <i>Orthopodomyia signifera</i> |
| 26. <i>Culex thriambus</i> | 56. <i>Psorophora ciliata</i> |
| 27. <i>Culiseta impatiens</i> | 57. <i>Psorophora columbiae</i> |
| 28. <i>Culiseta inornata</i>* | 58. <i>Psorophora ferox</i> |
| 29. <i>Culiseta melanura</i> | 59. <i>Psorophora howardii</i> |
| 30. <i>Culiseta morsitans</i> | 60. <i>Uranotaenia sapphi</i> |

Those in **bold** are species found Clark County, NV. Those * were found in wetlands during two year study. Courtesy of American Mosquito Control Association (AMCA) and Southern Nevada Health District (SNHD).

APPENDIX III

LIST OF PESTICIDES LABELED BY THE EPA FOR MOSQUITO CONTROL COLORADO STATE UNIVERSITY- ADULTICIDE AND PESTICIDE LIST

Adulticides

Product Name	EPA Registration Number	Company
ANVIL 10+ 10 ULV	1021-1688-8329	<u>Clarke mosquito control products</u>
ANVIL 2 + 2 ULV	1021-1687-8329	<u>Clarke mosquito control products</u>
BAYER AQUA- RESLIN	432-796	<u>Bayer Environmental Science</u>
BAYER PERMANONE 30-30	432-1235	<u>Bayer Environmental Science</u>
Bayer Pyrenone 25-5 public health	432-1050	<u>Bayer Environmental Science</u>
BIOMIST 1.5 + 7.5 ULV	8329-40	<u>Clarke mosquito control products</u>
BIOMIST 3 + 15 ULV	8329-33	<u>Clarke mosquito control products</u>
BIOMIST 30 + 30 ULV	8329-42	<u>Clarke mosquito control products</u>
CLARKE 5% SKEETER ABATE INSECTICIDE	8329-15	<u>Clarke mosquito control products</u>
CLARKE PERMETHRIN 57% OS	8329-44	<u>Clarke mosquito control products</u>
DIBROM 8 EMULSIVE NALED INSECTICIDE	5481-479	<u>Amvac Chemical Corporation</u>
DIBROM CONCENTRATE INSECTICIDE	5481-480	<u>Amvac Chemical Corporation</u>
FLIT 10 EC ULV	8329-69	<u>Clarke mosquito control products</u>
FYFANON	5905-196	<u>Helena chemical company</u>

Product Name	EPA Registration Number	Company
GRIFFIN ATRAPA ULV INSECTICIDE	1812-407	<u>Griffin LLC</u>
GRIFFIN ATRAPA VCP INSECTICIDE	1812-407	<u>Griffin LLC</u>
MALATHION 8 SPRAY	2935-83	<u>Wilbur-Ellis Company</u>
MASTERLINE AQUA-KONTROL CONCENTRATE	73748-1	<u>Univar USA, Inc.</u>
MASTERLINE KONTROL 2-2	73748-3	<u>Univar USA, Inc.</u>
MASTERLINE KONTROL 30-30	73748-5	<u>Univar USA, Inc.</u>
MASTERLINE KONTROL 4-4	73748-4	<u>Univar USA, Inc.</u>
MOSQUITOMIST 1.5 U.L.V.	8329-20	<u>Clarke mosquito control products</u>
PERMANONE 31-66	432-1250	<u>Bayer Environmental Science</u>
PERMANONE READY-TO-USE INSECTICIDE	432-1277	<u>Bayer Environmental Science</u>
PRENTOX PERM-X UL 30-30	655-811	<u>PRENTISS INCORPORATED</u>
PYROCID® Mosquito Adulticiding Concentrate	1021-1570	<u>McLaughlin Gormley King Company</u>
PYROCID® Mosquito Adulticiding Concentrate	1021-1569	<u>McLaughlin Gormley King Company</u>
SCOURGE INSECTICIDE	432-716	<u>Bayer Environmental Science</u>
TRUMPET EC INSECTICIDE	5481-481	<u>Amvac Chemical Corporation</u>
ULV MOSQUITO MASTER 412	8329-36	<u>Clarke mosquito control products</u>

Larvicides/ Pupicides

Product Name	EPA Registration Number	Company
1% SKEETER ABATE	8329-17	<u>Clarke mosquito control products</u>
5% SKEETER ABATE INSECTICIDE	8329-15	<u>Clarke mosquito control products</u>
ABATE 2-BG	8329-16	<u>Clarke mosquito control products</u>
ABATE 4-E INSECTICIDE	8329-60	<u>Clarke mosquito control products</u>
AGNIQUE MMF	53263-28	<u>COGNIS CORPORATION</u>
AGROSOLUTIONS AGNIQUE MMF, MOSQUITO LARVICIDE & PUPICIDE, MONOM	53263-28	<u>COGNIS CORPORATION</u>
AQUABAC 200G	62637-3	<u>BECKER MICROBIAL PRODUCTS, INC.</u>
AQUABACXT	62637-1	<u>BECKER MICROBIAL PRODUCTS, INC.</u>
BACTIMOS PELLETS FOR CONTROL OF MOSQUITO LARVAE	73049-51	<u>VALENT BIOSCIENCES CORPORATION</u>
BONIDE MOSQUITO LARVICIDE	4-195	<u>BONIDE PRODUCTS, INC.</u>
BVA 2 MOSQUITO LARVICIDE OIL	70589-1	<u>BVA INC</u>
CLARKE ABATE 1-BG	8329-17	<u>Clarke mosquito control products</u>
CONCENTRATE 1 A WATER EMULSIFIABLE INSECTICIDE CONCENTRATE	48665-2	<u>SHOO-FLY, INC</u>
GNATROL BIOLOGICAL LARVICIDE	73049-11	<u>VALENT BIOSCIENCES CORPORATION</u>
MOSQUITO DUNKS BIOLOGICAL MOSQUITO	6218-47	<u>SUMMIT CHEMICAL COMPANY</u>

Product Name	EPA Registration Number	Company
PRENTOX EMULSIFIABLE SPRAY CONCENTRATE #96	655-587	<u>PRENTISS INCORPORATED</u>
PRENTOX PYRONOL OIL CONCENTRATE OR-3610A	655-501	<u>PRENTISS INCORPORATED</u>
PRENTOX PYRONYL 303	655-797	<u>PRENTISS INCORPORATED</u>
PRENTOX PYRONYL CROP SPRAY	655-489	<u>PRENTISS INCORPORATED</u>
PRENTOX PYRONYL UL-100 CONCENTRATE	655-665	<u>PRENTISS INCORPORATED</u>
PRE-STRIKE	2724-451	<u>WELLMARK INTERNATIONAL</u>
STRIKE PROFESSIONAL MIDGE CONTROL	2724-446	<u>WELLMARK INTERNATIONAL</u>
SUMMIT B.T.I. BRIQUETS	6218-47	<u>SUMMIT CHEMICAL COMPANY</u>
TEKNAR CG	73049-403	<u>VALENT BIOSCIENCES CORPORATION</u>
TEKNAR G	73049-403	<u>VALENT BIOSCIENCES CORPORATION</u>
TEKNAR HP-D	73049-404	<u>VALENT BIOSCIENCES CORPORATION</u>
VECTOBAC 12AS BIOLOGICAL LARVICIDE AQUEOUS SUSPENSION	73049-38	<u>VALENT BIOSCIENCES CORPORATION</u>
VECTOBAC CG BIOLOGICAL LARVICIDE	73049-19	<u>VALENT BIOSCIENCES CORPORATION</u>
VECTOBAC G BIOLOGICAL LARVICIDE GRANULES	73049-10	<u>VALENT BIOSCIENCES CORPORATION</u>
VECTOLEX CG BIOLOGICAL LARVICIDE	73049-20	<u>VALENT BIOSCIENCES CORPORATION</u>
VECTOLEX WDG	73049-57	<u>VALENT</u>

Product Name	EPA Registration Number	Company <u>CORPORATION</u>
VECTOLEX WSP BIOLOGICAL LARVICIDE	73049-20	<u>VALENT BIOSCIENCES CORPORATION</u>
VET-KEM MOSQUITO LARVICIDE GRANULES SIPHOTROL	2724-451	<u>WELLMARK INTERNATIONAL</u>
ZODIAC PREVENTATIVE MOSQUITO CONTROL	2724-451	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID LIQUID LARVICIDE CONCENTRATE	2724-446	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID LIQUID LARVICIDE MOSQUITO GROWTH REGULATOR	2724-392	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID PELLETS MOSQUITO GROWTH REGULATOR	2724-448	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID SBG, SINGLE BROOD GRANULE, AN INSECT GROWTH REGULATOR	2724-489	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID XR EXTENDED	2724-421	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOSID XR-G	2724-451	<u>WELLMARK INTERNATIONAL</u>
ZOECON ALTOZID BRIQUETS	2724-375	<u>WELLMARK INTERNATIONAL</u>

APPENDIX IV

WETLAND DEVELOPMENT AND MANAGEMENT QUESTIONNAIRE- ADOPTED FROM MARIN AND SONOMA MOSQUITO AND VECTOR CONTROL DISTRICT

By answering the following questions, wetland developers and managers will have a better understanding of the concerns raised by local mosquito/vector control agencies when new or resurrected wetlands are proposed.

- Is adequate wetland drainage provided for?
- Are drainage facilities designed to drain both major and minor wetland areas and prevent ponding?
- Is the design of wetland basins adequate to minimize mosquito production?
- Are wetland management and maintenance provisions adequate?
- Is the probability of leakage or seepage from water conveyance systems and wetland basins considered?
- Will any abandoned wetland that could retain water be removed or reworked to prevent mosquito development satisfactorily?
- Are nearby industrial, commercial, suburban or urban operations identified?
- Is there adequate access and clearance for motorized mosquito control and wetland maintenance equipment?
- Do individual wetland basins have a drain to completely empty the structure, or can it be pumped dry adequately?
- If the wetland will hold waste water that is high in organic nutrients, i.e. animal or municipal waste water, has the greater potential for breeding mosquitoes been considered?
- If the project restores wetland habitat, has the probability of increased mosquito breeding (and higher adult mosquito populations) been considered?
- If the wetland will support mosquito fish for the control of mosquitoes, will the proper agency be notified prior to flooding of the wetland to ensure stocking of the fish?
- Have additional funds been reserved for the continual monitoring and control of mosquitoes if prevention standards are not heeded?
- Has the local mosquito control agency been informed of the plans?
- Has a mosquito management plan, surveillance plan, and control plan been established for the wetland?

APPENDIX V

ADVANTAGES AND DISADVANTAGES OF SURFACE FLOW AND SUBSURFACE FLOW WETLANDS.

Surface Flow (SSF) Wetlands	
Advantages	Disadvantages
Less expensive to construct (on a cost per acre basis) and operate and simpler to design than SSF wetlands and conventional treatment methods.	Lower rates of contaminant removal per unit of land than SSF wetlands, thus they require more land to achieve a particular level of treatment than SSF wetlands.
Can be used for higher suspended solids wastewaters.	Requires more land than conventional treatment methods.
More operating data in the United States than for SSF wetlands.	Risk of ecological or human exposure to surface-flowing wastewater.
Offer greater flow control than SSF wetlands	May be slower to provide treatment than conventional treatment
Offer more diverse wildlife habitat.	Odors and insects may be a problem due to the free water surface.
Provides habitat for plants and wildlife. Can offer natural mosquito control measures.	Higher mosquito populations
Subsurface Flow (SSF) Wetlands	
Advantages	Disadvantages
Higher rates of contaminant removal per unit of land than SF wetlands, thus they require less land to achieve a particular level of treatment than SF wetlands.	Requires more land than conventional treatment methods.
Lower total lifetime costs and capital costs than conventional treatment systems.	May be slower to provide treatment than conventional treatment
Less expensive to operate than SF systems.	More expensive to construct than SF wetlands on a cost per acre basis.
Minimal ecological risk due to absence of an exposure pathway.	Waters containing high suspended solids may cause plugging.
More accessible for maintenance because there is no standing water.	Plugging may result in outflow above surface allowing for mosquito outbreaks
Odors and insects not a problem because the water level is below the media surface.	Does not provide habitat for plants and wildlife.

APPENDIX VI

ESTIMATED MOSQUITO PRODUCTION PROPENSITY OF VARIOUS WETLAND PLANT SPECIES (COLLINS AND RESCH, 1989)

Plant group	Plant species	Common name	Mosquito production score
Rooted emergent plants	<i>Alisma geyeri</i>	Water-plantain	7
	<i>Alisma trivale</i>	Water-plantain	7
	<i>Alopecurus howellii</i>	Foxtail	9
	<i>Carex obnupta</i>	Sedge	11
	<i>Carex rostrata</i>	Sedge	14
	<i>Carex stipata</i>	Sedge	13
	<i>Cyperus aristatus</i>	Flat sedge	9
	<i>Cyperus difformis</i>	Flat sedge	11
	<i>Cyperus esculentus</i>	Flat sedge	13
	<i>Cyperus niger</i>	Flat sedge	12
	<i>Deschampsia danthonides</i>	Grass	11
	<i>Echinochloa crusgalli</i>	Barnyard grass	11
	<i>Echinodorus berteroi</i>	Burhead	10
	<i>Eleocharis palustris</i>	Spikerush	10
	<i>Equisetum arvense</i>	Horsetail	14
	<i>Frankenia grandifolia</i>	Alkali heath	14
	<i>Glyceria leptostachya</i>	Mannagrass	12
	<i>Juncus acutus</i>	Softrush	13
	<i>Juncus effusus</i>	Softrush	10
	<i>Jussiaea repens</i>	Primrose	16
	<i>Leersia oryzoides</i>	Rice cutgrass	11
	<i>Leptochloa fascicularis</i>	Salt-meadow grass	10
	<i>Ludwigia</i> spp.	Primrose willow	9
	<i>Lythrum californicum</i>	Loosestrife	13
	<i>Oryza sativa</i>	Rice	9
	<i>Phalaris arundinacea</i>	Reed canary grass	14
	<i>Phragmites communis</i>	Common reed	17
	<i>Plantago major</i>	Common plantain	9
	<i>Polygonum amphibium</i>	Water smartweed	14
	<i>Polygonum hydropiperoides</i>	Smartweed	12
	<i>Polygonum pennsylvanicum</i>	Pinkweed	12
	<i>Polygonum punctatum</i>	Smartweed	12
	<i>Polypogon elongatus</i>	Rabbitfoot grass	11
	<i>Potentilla palustris</i>	Cinquefoil	11
	<i>Pteridium aquilinum</i>	Fern	13
	<i>Sagittaria latifolia</i>	Duck-potato	7
	<i>Sagittaria longiloba</i>	Arrowhead	7
	<i>Sagittaria montevidensis</i>	Giant arrowhead	8
	<i>Scirpus acutus</i>	Bulrush	15
	<i>Scirpus americanus</i>	Three-square bulrush	10
	<i>Scirpus californicus</i>	Giant bulrush	15
	<i>Scirpus olneyi</i>	Alkali bulrush	12
	<i>Sparganium eurycarpum</i>	Burreed	13
	<i>Typha angustifolia</i>	Narrowleaf cattail	16
	<i>Typha glauca</i>	Cattail	16
	<i>Typha latifolia</i>	Common cattail	17
	<i>Zizania aquatica</i>	Wildrice	13

Plant Group	Plant Species	Common Name	Mosquito Production Score
Floating aquatic plants	<i>Azolla filiculoides</i>	Water fern	10
	<i>Bacopa nobisiana</i>	Water hyssop	13
	<i>Brasenia schreberi</i>	Water shield	12
	<i>Eichhornia crassipes</i>	Water hyacinth	18
	<i>Hydrocotyle ranunculoides</i>	Pennywort	15
	<i>Hydrocotyle umbellata</i>	Pennywort	15
	<i>Lemna gibba</i>	Duckweed	9
	<i>Lemna minima</i>	Duckweed	9
	<i>Nasturtium officinale</i>	Water cress	15
	<i>Nuphar polysepalum</i>	Spatterdock	11
	<i>Pistia stratiotes</i>	Water lettuce	18
	<i>Potamogeton crispus</i>	Curled pondweed	8
	<i>Potamogeton diversifolius</i>	Pondweed	8
	<i>Ranunculus aquatilis</i>	Buttercup	16
	<i>Ranunculus flammula</i>	Buttercup	15
	<i>Spirodela polyhyza</i>	Duckmeat	9
	<i>Wolffiella lingulata</i>	Bog mat	9
Submerged aquatic plants	<i>Callitriche longipedunculata</i>	Water starwort	11
	<i>Ceratophyllum demersum</i>	Coomtail	15
	<i>Eleocharis acicularis</i>	Spikerush	8
	<i>Elodea canadensis</i>	Waterweed	8
	<i>Elodea densa</i>	Waterweed	11
	<i>Isoetes howellii</i>	Quillwort	7
	<i>Isoetes orcuttii</i>	Quillwort	7
	<i>Lilaeopsis occidentalis</i>	Lilaeosis	7
	<i>Myriophyllum spicatum</i>	Water milfoil	14
	<i>Najas flexilis</i>	Naiad	11
	<i>Najas graminea</i>	Naiad	11
	<i>Potamogeton filiformis</i>	Pondweed	13
	<i>Potamogeton pectinatus</i>	Sago pondweed	13
	<i>Ruppia spiralis</i>	Ditchgrass	11
	<i>Utricularia gibba</i>	Bladderwort	12
	<i>Utricularia vulgaris</i>	Bladderwort	13
	<i>Zannichellia palustris</i>	Home d pondweed	10

APPENDIX VII

SNHD STANDARD OPERATING PROCEDURES VECTOR CONTROL- ENVIRONMENTAL HEALTH DIVISION ENCEPHALITIS VECTOR SURVEILLANCE (EVS) TRAP SET

Prep:

1. Traps use either 6V rechargeable or 3 D cell batteries. Use the volt meter to ensure batteries have adequate charge. For D cell batteries, discard if the volt meter reads less than 1.20V. 6V batteries should be fully charged.
2. Ensure each trap has a net, insulation in the bucket and that the fan/light works properly.
3. Each trap bucket uses 5 lbs of dry ice. Use the grey dry ice coolers for dry ice transport and ensure you have enough ice for all your traps.

Setting:

1. Due to the intense heat of the summer, set traps as late in the afternoon as possible.
2. Use the mosquito trap log (attached) and fill out all sections legibly.
3. Place a small paper tag with the trap number, date and location inside the net.
4. Place the trap between people and the source of the mosquitoes (pond, stream, woods, etc).
5. Place the traps where mosquitoes rest, near dense shrubs or bushes, but not directly in them, and not in high grass. Shaded areas are best. Avoid placing the trap in a sunny location. Choose a site that is protected from wind gusts and rain.
6. Place traps at shrubbery height, usually about 3 – 6 feet from the ground.
7. Turn trap light and fan on.

Pick up:

1. Due to the intense heat of the summer pick up traps as early in the morning as possible.
2. Put some dry ice in a cooler for storage and transportation of mosquitoes. Keep some plastic wrapping or burlap over the ice to provide a buffer between mosquitoes and dry ice.
3. Before turning off the trap, carefully and quickly remove the net and cinch it closed.
4. Turn off the trap and collect all parts.
5. Place the nets on top of the ice, ensuring there is a sufficient buffer between the mosquitoes and the ice.

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