Wyoming Health-Related Pest Control
Category 908-K
MOSQUITO CONTROL

University of Wyoming | Extension

Wyoming Health-Related Pest Control

Category 908-K

Mosquito Control

UNIVERSITY OF WYOMING | Extension

Issued in furtherance of extension work, acts of May 8 and June 30, 1914, in cooperation with the U.S. Department of Agriculture. Mandy Marney, director, University of Wyoming Extension, University of Wyoming, Laramie, Wyoming 82071. The University of Wyoming is an affirmative action/equal opportunity employer and institution and does not discriminate on the basis of race, color, religion, sex, national origin, disability, age, veteran status, sexual orientation, or political belief in any aspect of employment or services. The institution's educational programs, activities, and services offered to students and/or employees are administered on a nondiscriminatory basis subject to the provisions of all civil rights laws and statutes. Evidence of practices that are not consistent with this policy should be reported to the Employment Practices Office at (307) 766-6721.

Preparation for your Exam . . . . **v**

INTRODUCTION **1**

Chapter 1: DEVELOPMENTAL STAGES **2**

Learning objectives. 2
Egg. 3
Larva (plural: larvae) 4
Pupa (plural: pupae) 5
Adult 6

Chapter 2: ECOLOGY AND HABITAT **7**

Learning objectives. 7
Standing water mosquitoes 8
Permanent water group 8
Freshwater marsh. 8
Playa wetlands 9
Lakes 9
Ponds and seepage areas 9
Swamps 9
Transient (semi-permanent) water group 9
Borrow pits and canals 9
Freshwater drainage ditches 9
Floodwater group 10
Rain and floodwater pools 10
Wet meadows 10
Vernal pools 10
Artificial container and tree-hole group. 10
Tree holes 10
Artificial containers 10

Chapter 3: MOSQUITO SAMPLING AND SURVEILLANCE **12**

Learning objectives. 12
Mapping 13
Recordkeeping 14
Mosquito egg surveys 14
Sod sampling 14
Egg separation. 14
Oviposition trap 14
Larval and pupal surveillance. 15
Equipment 15
Dip procedure 15
Adult surveillance 16
Landing and biting collections 17
Insect sweep net collection 17
Bait trap collection. 17
Truck trap collection 17
Daytime resting collection 17
Light trap collection 18
Gravid trap collection 19
Other trap collection 19

Using mosquito survey data 19
Arbovirus surveillance in vectors and hosts 20
Communicating with other professionals 20
Sampling wild bird populations 21
Sentinel flocks 21
Arbovirus surveillance in mosquitoes 21
Transporting mosquitoes 22
Holding, identifying, and pooling collections 22

Chapter 4: CONTROL 23
Learning objectives 23
Source reduction 24
Sanitation 24
Water management 25
Biological control 25
Larvicidal applications 26
Larvicide application equipment 27
Spreader calibration 28
Power blower calibration 28
Adulticide applications 28
Adulticide application equipment 28
Barrier treatments 29
Space sprays 30
Resistance management 31
Genetic manipulation of mosquitoes 31

Chapter 5: PERSONAL PROTECTION AND EDUCATION 32
Learning objectives 32

Glossary 34

References 35
Table 1. Species of mosquitoes anticipated to occur in Wyoming (Darsie & Ward, 2005) 36

PREPARATION FOR YOUR EXAM

The reader preparing to take any of the Wyoming Commercial Pesticide Applicator Exam(s), including category 908-K, Mosquito Control, is encouraged to review the manual several times.

Exam questions can come from any portion of this manual.

It is important that you take note of the following:

- Exams are closed book. You will not be allowed to refer to any notes, manuals, or other unauthorized training materials during the exam.
- You may bring a basic handheld calculator with you to use during the exam (cell phones and other communication devices are prohibited—**you will be failed** if you use your cell phone during the exam).
- You must pass each category with a 70% or better to be issued a license.

Standards of Competence:

Applicators must demonstrate practical knowledge of appropriate life cycles and habitats of mosquito populations that form the basis of a control strategy. Applicators must demonstrate practical knowledge of a great variety of environments, ranging from exterior water sources to those conditions found in or on structures that promote mosquito populations, and have knowledge of non-chemical control methods such as sanitation, waste disposal, and drainage.

The information in this publication was obtained from the University of Florida, University of Minnesota, and the American Mosquito Control Association Public Health Pest Control website, which can be found at https://edis.ifas.ufl.edu/topics/vectors. This manual was revised by the Wyoming Mosquito Management Association Manual Revision Committee and Teton County Weed & Pest Control District.

Final edits by the University of Wyoming Pesticide Safety Education Program, 1/10/2025.

See references and/or hyperlinks for new content sources.
All uncredited photos by M. Iwaseczko.

INTRODUCTION

Mosquitoes are a type of fly and are scientifically classified in the order Diptera, meaning two-winged, and further classified as the family Culicidae. They are similar in body appearance to other flies but differ greatly in the specialized structure of their mouthparts. Mosquito mouthparts have evolved to pierce skin and extract blood from a variety of different hosts; because of this capacity, mosquitoes obtained their legendary bloodthirsty status. The family may be further divided into anophelines (which consists of *Anopheles* spp. only) and culicines, which contains all other mosquito genera occurring in Wyoming, including *Aedes*.

Table 1, (page 36) lists the 51 species of mosquitoes anticipated to occur in Wyoming.

Mosquitoes pass through four distinct stages in their life cycle: egg, larva, pupa, and adult. Due to spatial and temporal accumulations of mosquito eggs in some areas, impressive numbers of mosquitoes can hatch simultaneously under the proper conditions. In rapidly developing broods, survival of the immature stages can be quite high, but estimates for many species indicate that immature survival is normally less than 5 percent. Still, 5 percent of millions represents a sizable number. If they transmit disease or prefer to feed on humans, then they become appropriate targets for control activities. This manual provides an overview of mosquitoes' modes of life in their natural habitat and their adaptations to their surroundings; reviews surveillance practices; and outlines approved control methods that are applicable in Wyoming.

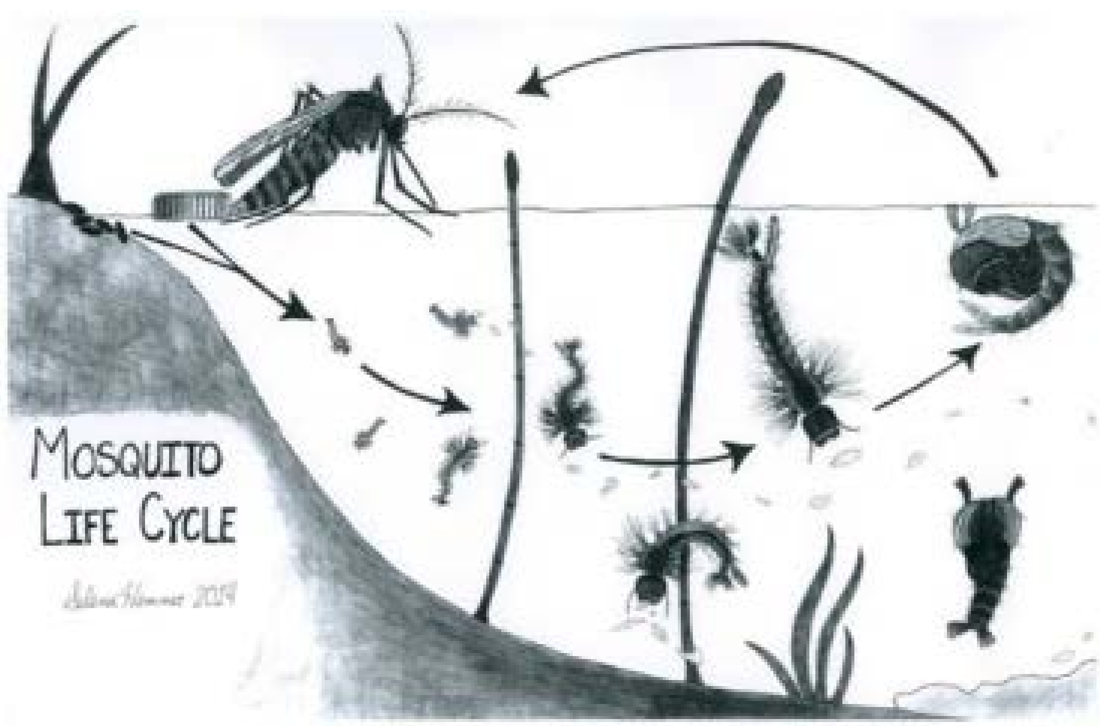


Figure 1. Mosquito life cycle.

CHAPTER 1: DEVELOPMENTAL STAGES

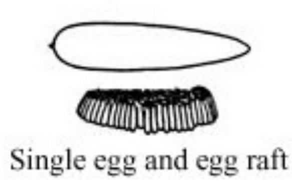
LEARNING OBJECTIVES

1. **Identify mosquito egg characteristics.**
   - Describe the initial and subsequent coloration of mosquito eggs.
   - Differentiate mosquito eggs based on their shape and the presence of specific structures such as floats in *Anopheles* species.
2. **Understand mosquito egg-laying behaviors.**
   - Compare and contrast the egg-laying behaviors of different mosquito species, including those that lay eggs singly versus those that form rafts.
   - Explain the importance of egg orientation in water for larval hatching.
3. **Analyze environmental influences on mosquito egg incubation.**
   - Discuss the environmental and genetic factors affecting the incubation period of mosquito eggs.
   - Describe how temperature, oxygen concentration, and photoperiod trigger egg hatching in different mosquito species.
4. **Describe mosquito larval development.**
   - Outline the four larval instar stages and the process of molting in mosquito larvae.
   - Identify key morphological features of mosquito larvae, including the siphon, and explain their function.
5. **Examine mosquito pupation and emergence.**
   - Explain the characteristics and behaviors of mosquito pupae.
   - Describe the process of eclosion and the vulnerabilities associated with this stage.
6. **Differentiate adult mosquito characteristics and behaviors.**
   - Distinguish between male and female adult mosquitoes based on morphological features and feeding behaviors.
   - Explain the role of female mosquitoes in disease transmission and the factors influencing their lifespan and reproductive cycle.

EGG

Mosquito eggs are white in color when first deposited but darken within 12 to 24 hours. Most species' eggs appear similar when seen by the naked eye, except for those of the *Anopheles* spp., whose eggs have floats attached to each side. When viewed with magnification, eggs of different species can be seen to vary from canoe shaped to elongate or elongate-oval in shape and this, coupled with other characteristics, may be used to identify eggs to species. Myers (1967) created an identification key to mosquito eggs that is applicable to many of the *Aedes* species commonly found in Wyoming.

Figure 2. Egg raft dimple.



Most mosquito species lay eggs singly while others, including most *Culex*, *Coquillettidia*, and *Culiseta* species glue them together to form rafts. These rafts can be easily spotted on the surface of the water due to a dimple formed from the interaction of a lipid (fatty substance) portion of the eggs that keeps the raft upright in the water. This orientation is critical for the larvae to be able to hatch into water (Clements, 2008).

The incubation period, or elapsed time between **oviposition** (egg deposition) and readiness to hatch, is dependent on environmental and genetic factors and varies considerably among different species.

Figure 3. Mosquito hatch.

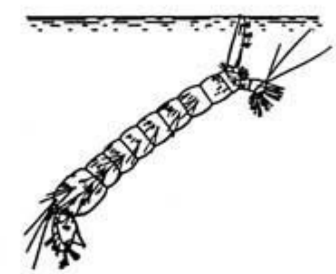
Permanent standing water (*Culex*, *Culiseta*, and *Anopheles*) species deposit their eggs directly on the water's surface, and these may hatch in one to four days depending on temperature. Many floodwater and container-breeding species deposit their eggs on moist soil or other wet substrates. These eggs may hatch within a few days after being flooded, or the fully developed larvae may remain within the eggs for up to a year or more. These eggs often require desiccation or frost conditioning, and then hatch depending on immersion conditions.

In early spring, upon flooding, the hatching trigger for these eggs is often dissolved oxygen concentration. A decrease of oxygen in the water column indicates that organic material is decomposing, or bacteria are proliferating and respiring, and that a plentiful food source is available for the newly hatched larvae to eat. In late spring and summer, the hatching trigger is often a rise in water temperature (likely also an indication of increased biological activity in the immediate environment) or some combination of temperature, oxygen concentration, and **photoperiod** (day length). The dormant eggs laid in floodplain depressions or in an abandoned tire accumulate over time, due to continued oviposition by blood-fed females. When temporarily flooded, they hatch along with more recently deposited eggs. Resulting larval populations can attain large numbers quickly this way.

LARVA (PLURAL: LARVAE)

The larvae (also known as wigglers or wrigglers) of all mosquitoes live in water. They undergo four stages of growth, called instars. Upon each instar they molt, or shed their previous exoskeleton, to accommodate subsequent growth. First instar larvae hatch from mosquito eggs using a hard plate on their head called an **egg-burster** and can be distinguished under magnification by this tooth-like structure on the top of their head.

Subsequent larval instars can be visually gauged by the relative size of the larva. However, it is important to note that there is variability in the size of different species. For instance, the small fourth instar of *Aedes cinereus* can easily be mistaken for a third instar of *Aedes vexans* or even a second instar of *Culiseta inornata*. To further complicate identification, crowding or a lack of food may decrease the size of the larvae. As will be discussed later, **being able to discern instar is critical for effective control of mosquito larvae.** This requires familiarity with the species known to develop in the habitat being surveyed.



Nonanopheline larva

Another structure used for identification is the siphon. Near the last abdominal segment in most species is a siphon, or air tube, that serves as a breathing apparatus when the larva hangs down vertically below the water's surface. The larva uses the opening at the tip of the siphon to breathe atmospheric air. The size and shape of this siphon can help determine the genus of the larvae at hand. Generally, the siphons of *Culex* spp. are long and slender compared to those of *Aedes* spp., whereas *Psorophora* siphons tend to appear bulbous. Larvae of *Anopheles*, however, lack this appendage, and breathe through a cluster of small abdominal plates, which prompts them to lie flat close to the underside of the water's surface when not diving. *Coquillettidia perturbans* larvae on the other hand, have a highly specialized siphon that attaches to emergent vegetation. Serrations and a curved tooth on the siphon enable larvae to burrow into and securely attach to a cattail stem, utilizing the plant's interior airway for their oxygenation needs (Bosak & Crans, 2002). **Their resulting absence from the water column and surface renders traditional sampling methods ineffective at determining larval densities of *Cq. perturbans***

Larva of *Anopheles* spp.

and necessitates using a modified bilge pump (Walker & Crans, 1986) to sample the substrate at the roots of the cattail plants.

Finally, early instar larvae of most species can acquire adequate oxygenation through passive diffusion of oxygen through their cuticle (skin) and consequently may not be easily sampled from the water's surface (Clements, 2008). Preferential sampling *within* the water column and a keen eye can help detect their subtle but often considerable presence.

The larvae of certain mosquito species (e.g., *Psorophora ciliata*) are predaceous and prey on other invertebrates, including other mosquito larvae. Most larvae though, are indiscriminate filter feeders, ingesting anything smaller than about 10 microns by vibrating their mouth brushes and sweeping in suspended matter. Organic particles, such as pollen deposited on the water, and micro-organisms, such as bacteria, viruses, protozoans, and fungi, are filtered from the surrounding water. The vibration of the mouth brushes can also serve to propel mosquito larvae through the water column. Later instars of several species of mosquitoes will also scrape or graze on algae or bacteria on submerged surfaces (Clements, 2008).

Depending on the species and environmental conditions, particularly temperature, mosquitoes may take anywhere from three to four days up to several weeks to complete larval development. The latter is especially true in Wyoming springtime, where low nighttime temperatures can keep water temperatures and larval development to a minimum.

Mature fourth instar larvae molt to the pupal stage. Prior to that, the larvae stop feeding and begin development of pupal structures, including the breathing trumpets, which may occasionally be visible under the larval cuticle.

PUPA (PLURAL: PUPAE)

Pupa is the stage in an insect's life where it transforms from a larva to an adult. Unlike most other insects, mosquito pupae can be very active and are often called "**tumblers**" because of their rapid, tumbling-like movement when disturbed. Mosquito pupae breathe through two respiratory "horns" when at the water's surface; they do not feed and are propelled through the water column by rear paddles. Pupae typically transform into adults in two or more days. When emergence nears, the pupae spend more time near the water's surface and begin to straighten out.

Pupa

During **eclosion** (emergence from the pupal skin), the pupa breathes in excess atmospheric air, which splits open a seam on its back. The adult mosquito then pushes upward like a folded tripod and, when completely free of the pupal case, spreads its legs on the water's surface. This is a particularly vulnerable time for the mosquito, as it is subject to potentially lethal wave action and wind, and underscores the critical importance of the female's choice of oviposition sites. The adult mosquito then waits several minutes to dry and harden, then flies away.

ADULT

All adult mosquitoes are capable of flight. Male mosquitoes are the first to emerge from a brood because they require time to mature sexually prior to the females' emergence. This is of great benefit to mosquito controllers, as a misinterpretation of larval instar or an emergence of male mosquitoes can still be quickly corrected with a **pupicide** application.

Adult male mosquitoes can be easily distinguished from females by their feathery antennae. These antennae have dense bristles used to locate females of their species for mating through their recognition of distinct wingbeat frequencies. Male mosquito mouthparts are modified to probe flowers for nectar and plant secretions. Females also feed on plant nectar for everyday energy needs.

While shaped similarly to their male counterparts, female mosquitoes' piercing-sucking mouthparts are significantly more robust. The six needle-like stylets enable the mated females to penetrate the skin of their preferred host (animal or bird) for a blood meal, a source of protein necessary for most species in the production of eggs. While the female sucks in blood, she simultaneously injects saliva into her host to facilitate her meal. It is in this process that a mosquito may spread a pathogen from one host to the next.

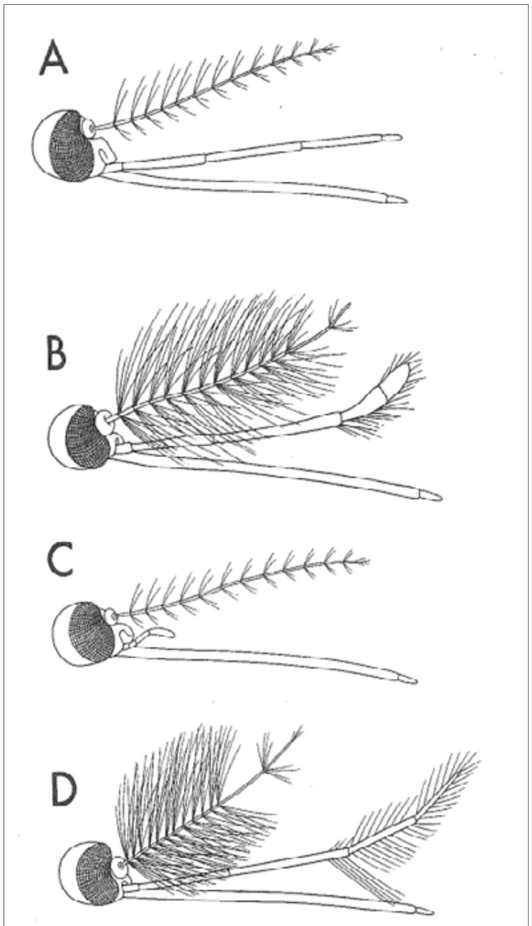


Fig. 4. Head and appendages of mosquitoes. *A*, Female *Anopheles*. *B*, Male *Anopheles*. *C*, Female Culicine. *D*, Male Culicine.

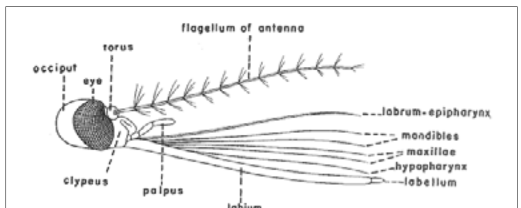


Fig. 5. Mouth parts of female mosquito.

Eggs develop within the female a few days after she takes a blood meal. Females oviposit on the water, in crevices in the soil, on other favored substrates, or special niches that are or will subsequently be flooded, such as natural and artificial containers or tree holes; then the life cycle repeats itself. The adults of some species remain within a few hundred feet of where they spent the larval stage, while others may migrate up to 50 miles or more. Females of some floodwater species may live up to a month after they emerge; some permanent water or standing water species can survive for several months by overwintering as mated adults. Some species, including those whose eggs require freezing temperatures, are limited to a single generation per year, whereas others have multiple generations per year.

CHAPTER 2: ECOLOGY AND HABITAT

LEARNING OBJECTIVES

1. **Differentiate mosquito species.**
    - Identify morphological differences among mosquito species in larval and adult stages.
    - Recognize how these differences aid in accurate species identification.
2. **Understand mosquito behavioral and habitat differences.**
    - Explain how behavioral differences allow mosquito species to occupy various ecological niches.
    - Describe the significance of identifying breeding habitats for mosquito control.
3. **Plan mosquito control strategies.**
    - Discuss the importance of knowing mosquito species' behaviors and habitats in developing effective control strategies.
    - Identify the steps involved in the identification of problem species and their breeding habitats.
4. **Analyze aquatic environments for mosquito breeding.**
    - Compare different aquatic environments where mosquito larvae and pupae can develop.
    - Assess how water chemistry, vegetation, and sunlight exposure influence mosquito breeding sites.
5. **Identify standing water mosquito habitats.**
    - Describe the characteristics of standing water habitats such as freshwater marshes, playa wetlands, lakes, ponds, and seepage areas.
    - List mosquito species commonly found in each type of standing water habitat.
6. **Understand floodwater mosquito habitats.**
    - Explain the breeding habits of floodwater mosquito species and their reliance on periodic flooding.
    - Identify typical floodwater mosquito habitats such as rain pools, wet meadows, and vernal pools.
7. **Recognize transient water mosquito habitats.**
    - Describe the features of transient water habitats like borrow pits, canals, and freshwater drainage ditches.
    - Identify mosquito species associated with these transient water habitats.
8. **Identify artificial container and tree hole habitats.**
    - Explain how tree holes and artificial containers serve as breeding sites for certain mosquito species.
    - List species commonly found in these habitats and the diseases they may transmit.
9. **Correlate habitat preferences with mosquito surveillance.**
    - Discuss how knowledge of habitat preferences can enhance mosquito surveillance and control efforts.
    - Explain the challenges faced by control agencies when dealing with mosquitoes that thrive in diverse habitats.
10. **Assess environmental impacts on mosquito life cycles.**
    - Evaluate how environmental conditions, such as seasonal changes and water availability, affect mosquito life cycles and population dynamics.
    - Explain the concept of diapause and its role in mosquito survival during harsh conditions.

Those casually aware of mosquitoes may believe that all types are much the same, and, indeed, the similarities between species are considerable. There are, however, many differences in appearance from species to species and even among some varieties within species. These morphological differences, especially notable in the larval and adult stages, permit accurate identification of most species. Behavioral differences permit various species to occupy numerous ecological niches with relatively little overlap. Thus, knowledge of the source or breeding habitat of mosquitoes can provide strong clues to their identification.

Planning and carrying out a treatment plan requires knowledge of the behavioral and habitat differences among mosquito species. The trained worker first identifies the problem species. Once identity is established, useful correlations are immediately available, such as the type of breeding habitat and where to search for larvae. A working knowledge of the behavior and habitats frequented by various species aids in determining the kinds of survey and control strategies best suited for the task.

All mosquitoes are aquatic in the first (larval and pupal) stages of life. Though not adapted to life in moving waters, some can occupy the quiet pools and seepage areas near flowing streams and just about any other standing water source. Aquatic environments differ largely in the chemistry of the water (acidic or alkaline; fresh, salt, or brackish). These environments may be natural or man-made and may also differ in the amount or type of vegetation present and the amount of sun or shade. Noting these variations, whether formally or informally, can improve predictability by associating habitat preferences with local species, and renders future surveillance and treatment efforts more effective.

Although some species use more than one type of habitat, most mosquitoes can be categorized in general terms by their preference for either **standing water** sites or **floodwater** sites.

STANDING WATER MOSQUITOES

Standing water species deposit their eggs either singly or in rafts on the surface of permanent or semi-permanent pools of standing water. They usually produce several generations (broods) each year and overwinter or survive harsh environmental circumstances as mated females. Knight et al. (2014) cites a scant 5% of Wyoming wetlands that remain flooded year-round and nearly two-thirds dry out by mid to late summer each year. These conditions impact the life cycle of species that depend upon standing water, prompting fewer generations per season and/or earlier winter suspended development. This resting/suspended development period is called **diapause.**

PERMANENT WATER GROUP

Mosquito groups assigned to the permanent water group are *Anopheles* spp., *Culex* spp., *Coquillettidia perturbans,* and *Uranotaenia sappharina*.

Freshwater marsh

According to Knight et al. (2014), in the mountains of Wyoming, freshwater marshes occur in glacial potholes, old beaver ponds, and abandoned stream channels. Associated plant communities include Northwest Territory sedge and water sedge, twinleaf bedstraw, elephant head lousewort, white marsh marigold, and pond lily. Common willow species include Booth's, diamond leaf, Drummond's, Geyer's, and Wolf's willows. At lower elevation, marshes occur mostly in abandoned stream channels and in irrigation backwaters. Dominant plant species include Nebraska sedge, cattail, common spikerush, threesquare bulrush, hardstem bulrush, arumleaf arrowhead, and water knotweed. Mosquito species often found in freshwater marshes include *An. walkeri, Cx. salinarius, Cq. Perturbans,* and *Cx. tarsalis.*

Playa wetlands

According to Tibbetts et al. (2016), "Barren and sparsely vegetated playas [have] generally <10% plant cover. Salt crusts are common in playas, small salt grass beds are often established in depressions, and sparse shrubs grow around the margins. These systems flood intermittently. Impermeable alkaline to saline clay hardpans preclude surface water from infiltrating and standing water eventually evaporates. Salt encrustations can occur on the surface. Salinity varies greatly depending on soil moisture and has a major influence on species composition. Characteristic vegetation includes greasewood (*Sarobatus vermiculatus*) and salt grass (*Distichlis spicata*)." The mosquito species most likely to occur in this habitat include *Cx. tarsalis*, *Ae. nigromaculis*, *Psorophora signipennis*, and *Ae. vexans.* Large numbers of *Aedes* and *Psorophora* can be produced following the heavy rains of June or July, followed by increasing numbers of *Culex* as the emergent annual vegetation increases in the playa (Rey et al., 2012).

Lakes

Larvae may be found when many species of floating or emergent plants are present. When vegetation occurs only in a narrow band along the lakeshore, larvae are confined to this littoral zone. Lake species include *An. walkeri, Uranotaenia sappharina, Cx. salinarius,* and *Cq. perturbans.*

Ponds and seepage areas

There is no clear distinction between a pond and a lake except that ponds are generally smaller. Grassy woodland ponds or fluctuating ponds occupy shallow depressions and are filled by rainwater or surface runoff. They are usually of uniform depth, but the area they cover varies, depending on rainfall. Sinkhole ponds are usually quite deep and may be covered with vegetation, or free of all except marginal plants. Both types of ponds may contain larvae of *An. earlei, Culiseta inornata, Cx. restuans, Cx. salinarius, Cx. territans,* and *Aedes canadensis.* The seepage areas around hillsides and ponds or streams most often breed *An. freeborni* and *Ae. sticticus.*

Swamps

Swamps differ from marshes principally in having dense cover from larger trees. The most common species of mosquito larvae found here are *Ae. canadensis* and *Cq. perturbans.*

TRANSIENT (SEMI-PERMANENT) WATER GROUP

Mosquito groups assigned nationally to the transient water group are *Cx. tarsalis, Cx. restuans,* and *Cs. inornata.*

Borrow pits and canals

These man-made bodies of open water produce more mosquitoes as they silt in and become overgrown with vegetation. They yield *An. punctipennis, Cs. inornata, Ae. canadensis, Cx. nigripalpus, Cx. restuans, Cx. salinarius,* and *Cq. perturbans.*

Freshwater drainage ditches

In pastures, at the bottom of road shoulders, in old fields, and in lowland groves, freshwater ditches will often yield the following species of mosquito larvae: *Cx. nigripalpus, An. walkeri,* and *Ur. sapphirina.*

FLOODWATER GROUP

Floodwater species deposit their eggs in locations subject to periodic flooding, such as damp soil in depressions or inside tree holes and artificial containers. They produce one to several broods annually, and overwinter or survive harsh environmental circumstances in the egg stage. Mosquitoes assigned nationally to this floodwater group are *Ae. dorsalis, Ae. nigromaculis,* and *Ae. vexans.*

Rain and floodwater pools

These pools form the breeding place for many species, especially *Psorophora* and *Aedes.* The pools disappear in dry weather and support no true aquatic vegetation, though usually a layer of leaves and other detritus settles on the bottom. Mosquito species found in this habitat are *Ae. increpitus, Ae. trivittatus, Ae. idahoensis, Ae. sticticus, Ae. vexans,* and *Ae. cinereus.* Rees and Nielson (1951) found *Ae. implicatus* associated with streamside pools in willow habitat at elevations between 6,000 and 9,000 feet.

Wet meadows

Wet meadows are herbaceous wetlands associated with a high water table or overland flow, but typically lack standing water. Sites with no channel formation are typically associated with snowmelt or groundwater and not subjected to high disturbance events such as flooding. Sites associated with a stream channel are more tightly connected to overbank flooding from the stream channel than with snowmelt and groundwater discharge. Vegetation is dominated by herbaceous species; typically, graminoids, or grass-like plants, have the highest canopy cover including *Carex* spp. (sedges), *Calamagrostis* spp. (reed grasses), and *Deschampsia caespitosa* (tufted hair grasses) (Tibbets et al. 2016). Mosquitoes associated with this habitat include *Ae. campestris*, *Ae. dorsalis*, *Ae. flavescens*, *Ae. vexans*, *An. earlei*, *Cx. tarsalis*, *Cx. territans,* and *Cu. inornata*.

Vernal pools

These pools flood seasonally with rainfall, snowmelt, or runoff and are usually dry in summer and fall; vegetation is highly variable. Fairy shrimp and a lack of fish are good indicators of this habitat, which retains water for a longer period than surrounding areas due to an impermeable layer of soil below. *Ae. canadensis* and *Ae. excrucians* are associated with this habitat type.

ARTIFICIAL CONTAINER AND TREE-HOLE GROUP

Tree holes

Tree holes or rotten tree cavities support a rather unusual mosquito fauna, with many species breeding almost exclusively in this habitat. Resident species are *Ae. sierrensis* and *Ae. hendersoni.* Lunt and Peters (1976) found *Ae. hendersoni* associated with cottonwood, American elm, willow, hackberry, boxelder, green ash, and white mulberry. *Ae. sierrensis,* though currently confined geographically to the western portion of Wyoming, is a species to note due to its vectorial capacity for dog heartworm. In California, Huang et al. (2013) found that in the absence of the primary vector (*Ae. sierrensis*), *Cx. pipiens*, *Cx. tarsalis, Ae. melanimon, Cu. incidens,* and *Cu. inornata*—all species found in Wyoming—demonstrated a high field infection rate with heartworm.

Artificial containers

Several species breed in human-created situations around human dwellings. Horse troughs, fish pools, cisterns, rain barrels, rain gutters, tarp depressions, and old tires, etc., containing water serve as excellent larval habitat. Species most often encountered are *Cu. inornata*, *Cx. restuans, Cx. pipiens,* and *Cx. salinarius.* Introduced container-breeding species that are spreading across North America include *Ae. japonicus, Ae. aegypti,* and *Ae. albopictus.* Due to the capacity of the latter two species to transmit a multitude of diseases, including yellow fever, dengue, and Chikungunya and now Zika viruses, vigilance for the presence of these species is of utmost importance.

While habitat association with many species is quite specific, others thrive in a variety of situations. Thus, the detection of adults of these latter species in routine surveys does not provide an immediate indication of the related breeding site(s) and adds to the complexity of control.

CHAPTER 3: MOSQUITO SAMPLING AND SURVEILLANCE

LEARNING OBJECTIVES

1. **Identify mosquito species.** Understand the methods for identifying different mosquito species and their relevance to disease transmission and nuisance control.

2. **Conduct initial surveys.** Learn how to perform initial surveys to gather information on mosquito species, locations, densities, and disease potential in a given area.

3. **Implement surveillance programs.** Develop skills in establishing and conducting routine mosquito surveillance programs, including monitoring adult and larval populations, rainfall, runoff, and breeding site locations.

4. **Mapping for mosquito control.** Gain proficiency in using and creating accurate and comprehensive maps for mosquito control operations, including the use of GIS technology and GPS coordinates for precise location tracking.

5. **Recordkeeping and data management.** Understand the importance of consistent and accurate recordkeeping in mosquito surveillance and control, including the use of standardized forms and data-recording devices.

6. **Mosquito egg surveys.** Learn techniques for conducting mosquito egg surveys, including sod sampling and egg separation, to determine exact breeding locations and species present.

7. **Larval and pupal surveillance.** Acquire skills in larval and pupal surveillance, including the use of specialized equipment and techniques to locate breeding sites and estimate mosquito populations.

8. **Adult mosquito surveillance.** Master various methods of adult mosquito surveillance, such as landing and biting collections, light trap collections, and bait trap collections, to assess mosquito incidence and abundance.

9. **Use surveillance data.** Understand how to use mosquito survey data to make informed decisions about control measures, evaluate the effectiveness of control operations, and correlate mosquito populations with disease prevalence and nuisance levels.

10. **Arbovirus surveillance.** Learn the methods for arbovirus surveillance in mosquito populations and hosts, including communication with healthcare and veterinary professionals, sampling wild bird populations, and using lab-based testing methods like ELISA and PCR.

11. **Field operations.** Develop practical skills in field operations, including recognizing mosquito breeding behaviors, using dip procedures for larvae collection, and differentiating mosquito larvae from other aquatic insects.

12. **Effective communication and coordination.** Understand the importance of communication and coordination with public health agencies, local healthcare practitioners, and veterinary laboratories for effective mosquito-borne disease surveillance and control.

Surveys are essential for the planning, operation, and evaluation of an effective mosquito control program. These programs exist to prevent mosquito-borne disease and/or reduce nuisance mosquito populations to socially acceptable levels. Initial surveys identify the species of mosquitoes present and provide general information on locations, densities, and disease potential. With this knowledge, it may be possible to determine life cycles and feeding preferences; predict larval habitats, adult resting places, and flight ranges; and perhaps even make preliminary recommendations for control programs.

The next step is to embark on a formal surveillance program in which routine monitoring of mosquito presence is conducted. A basic inspection program usually addresses adult and larval population density and species composition, rainfall/runoff monitoring, and breeding site locations. Additional specialized surveillance may be conducted to detect arboviral presence in birds and mosquito populations, operation of **ovitraps** (e.g., for *Ae. aegypti* and *Ae. albopictus* surveillance), or sampling of floodwater mosquito eggs to locate breeding sites. This information not only provides justification for source reduction and insecticide applications, but it also serves as an ongoing indicator of the effectiveness of these activities and continually adds to the database of knowledge concerning mosquitoes in the area. Such inspections do not determine the absolute population of mosquitoes, but they can show fluctuations in relative mosquito abundance and diversity over time in the various habitats visited.

MAPPING

Reasonably accurate and comprehensive maps are essential in conducting a mosquito control operation. Maps provide information for field survey and control activities, program evaluation, and reporting and budgeting. They show elevations, streets, roads, and railroads, as well as ponds, lakes, streams, sewage lagoons, flooded fields, and other breeding areas. They are used for orientation and for locating and plotting larval breeding places and adult sampling stations. When large areas are involved, a master map may be needed for planning drainage and other field operations. The master map will indicate the treatment areas, the possible flight range of mosquitoes from breeding sites and, with prevailing wind direction, the potential degree of penetration into populated areas. Larval and adult sampling stations can be indicated by symbols and numbers. Counts made at these stations at weekly or biweekly intervals provide information for current evaluation of the mosquito problem by indicating the abundance of mosquitoes, species involved, flight range, habitat, and disease potential. This information identifies areas requiring high priority for treatment. Narrative descriptions, sometimes necessary for exact location description, are simplified whenever possible. For example, "N.W. corner of 15th Street and Ninth Avenue" is a brief description that leaves no doubt as to the location. There may be some areas that are difficult to accurately locate (e.g., marshlands). However, maps can be subdivided into numbered or named areas for easy reference, and Global Positioning System (GPS) coordinates are very reliable. Some common methods of subdividing maps involve the use of geographical features (such as different habitats or fenced fields), artificial grids, or a combination of both to set boundaries on areas that are indexed for easy reference and filing. To avoid cluttering, the larger areas may be further subdivided using transparent overlays, again employing geographical features or a grid. Once the area of inspection is delineated by reference to index numbers, additional location data can be conveyed clearly using cards that include a rough sketch of the area or incorporated into a Geographic Information System (GIS) format. Many local government agencies and districts have the software to produce and manipulate maps and use resources such as recent aerial imagery (available at USDA's NRCS website: https://gdg.sc.egov.usda.gov). This enables the collection of multiple data types and sets with the ability to view them spatially on a map.

RECORDKEEPING

To avoid comparing dissimilar parameters, inspections should be consistent both in method and location. Keeping clear, accurate records is as important as the data gathering itself. Surveillance records are systematically managed to ensure that subsequent inspections can be accurately conducted by others less familiar with the area. They usually include the inspector's name, date of inspection, and exact location, in addition to the mosquito-related data collected.

Data-recording forms and devices promote uniformity. These make records easier to read, interpret, and summarize, and serve as a reminder to the inspector to record all pertinent information. In the absence of data recorders, standardized formats lead to more consistently accurate transcription of the data into permanent records.

MOSQUITO EGG SURVEYS

Egg surveys are carried out primarily to determine the exact breeding locations of mosquitoes. *Aedes, Ochlerotatus,* and *Psorophora* mosquitoes lay their eggs on damp soil in places subject to intermittent flooding. Two types of egg surveys can be conducted for these genera: sod sampling and egg separation.

Sod sampling

Sod samples, usually containing 8 cubic inches of soil and vegetation with a thickness of about an inch, are stored for a week or more to allow the embryos time to develop within the eggs. The sod samples are then placed in glass jars and flooded with water. The larvae are identified after they hatch. Several sequential flooding and drying periods may be necessary to get sufficient cumulative hatch. In larval surveys, sod sampling delineates breeding areas, especially when sampling is done during times when larvae are not present.

Egg separation

Egg separation machines can be used for separating mosquito eggs from soil and debris by mechanical agitation, washing, screening, or sedimentation of debris and flotation of the eggs in a saturated salt solution. Sod or soil samples are cut in the field with a sharp trowel around a 6-inch-square template, placed in plastic bags, and stored (sometimes for months) in a cool room. The various species and densities of *Aedes, Ochlerotatus,* and *Psorophora* can be identified by microscopic examination of live or preserved eggs using taxonomic keys for mosquito eggs.

OVIPOSITION TRAP

Collections of mosquito eggs in oviposition traps are used to detect and monitor container-breeding mosquitoes such as *Ae. triseriatus, Ae. sierrensis,* *Ae. aegypti* , *Ae. japonicus,* and *Ae. albopictus.* The oviposition trap can easily be made of plastic stadium cups or cemetery urns, food cans (3-pound coffee cans), or pint jars painted black inside and outside. The traps are placed in shaded areas at a height no greater than 4 feet and filled with water and a few dried leaves placed at the bottom of the container. An oviposition substrate made of a strip of various materials (seed germination paper, muslin, formica, balsa wood, wooden tongue depressor, etc.) is then placed vertically inside the container with the water covering about half of it. Gravid females use this substrate to lay eggs just above the water level. Traps are checked every 10 to 14 days to prevent them from becoming breeding sources. If larvae are found in the trap, then the water should be emptied, and the trap reset. The ovipositional substrate is periodically collected and returned to the laboratory in a plastic bag. Samples are kept cool and moist during transportation, taking care to avoid too much moisture, which could cause eggs to begin hatching. Eggs, or the resulting fourth instar larvae, are then identified.

LARVAL AND PUPAL SURVEILLANCE

Before beginning a survey, obtain information about the general breeding behavior and habitats of the species known or suspected to be in the area. An experienced person may be able to spot the probable mosquito breeding places in a specific area by rapid reconnaissance. These areas are carefully numbered and marked on the map. Determining the specific breeding sites and establishing permanent larval sampling stations requires a more detailed inspection. Larval surveys help determine the exact areas in which the mosquitoes breed and their relative abundance. This information is valuable for all control operations.

EQUIPMENT

A white enameled or plastic dipper about 4 inches in diameter (1 pint or 350 mL capacity) is frequently used for collecting mosquito larvae. The handle of the dipper may be lengthened by inserting a suitable piece of wood dowel or PVC pipe. Specially designed dippers can be created so that their capacity can be directly related to the amount of water surface examined. Thus, the number of larvae per square foot or meter can be computed with reasonable accuracy.

DIP PROCEDURE

Mosquito larvae of some species are usually found near surface vegetation or debris, which provide the larvae with both food and cover from predators. In larger ponds or bodies of water, these larvae are typically confined to the calmer shoreline areas. It is necessary to proceed slowly and carefully in searching for mosquito larvae as disturbance of the water or shadows may cause the larvae to dive to the bottom.

Anopheline larvae (which lie parallel to the water's surface) are collected by a skimming movement of the dipper with one side pressed just below the surface. The stroke ends just before the dipper is full because larvae will be lost if the dipper is filled to the point that it runs over. Where clumps of erect vegetation are present, it is best to press the dipper into a clump with one edge depressed so that the water flows from the vegetation into the dipper. **Culicine** larvae such as *Ae. vexans, Ae. idahoensis,* or species of *Psorophora* require a quicker chopping motion of the dipper as they are more likely to dive below the surface when disturbed.

Certain species are able to remain submerged for extensive periods of time. For example, Means (1981) reported *Ae. dorsalis* could remain submerged for over 5 minutes. This underscores the need to both be aware of the behaviors of the species inhabiting each sampling location and to approach the habitat carefully to avoid spooking the larvae. In deeper water, the dipper may be used to scout for larvae by creating a light-colored background to facilitate spotting the larvae within the water column. It is also important to note that water temperature affects the mobility of the larvae. On early spring mornings in higher elevations, colder water temperatures cause the larvae to move only if prodded. This may lessen the value of using movement to detect larvae in a dipper and may force the inspector to rely on recognizing the shape of larvae floating in the water. Occasionally, an inspector may encounter what appears to be the ghosts of larvae floating in the water. These represent the **exuviae**, or shed skins, of the larvae or pupae that have moved on to their next stage of development (and can leave an inspector truly disheartened).

Differentiating mosquito larvae from other aquatic insects is of great importance in effective surveillance and control. A variety of midges can be mistaken for either the shape or movement of mosquitoes in the field. For images and videos to enable this differentiation, see the Wyoming Mosquito Management Association website (https://sites.google.com/view/wyomosquito).

The inspector records the number of dips made and the number of larvae found, by **instar** if warranted, and transfers representative sample specimens by pipette into small vials of alcohol for later identification. With most species, it is possible to get a rough idea of the breeding activity by computing the average number of larvae of each species per dip. The number of dips required will depend on the size of the area and the relative larval density, but for convenience is often in multiples of 10. Inspection should be made at weekly or biweekly intervals during the mosquito breeding season, as areas that are entirely negative at one time may rapidly become heavily infested. Inspections for certain species require variations in the procedure described above. For example, *Coquillettidia* larvae remain below the surface throughout much of their development, attached by the **siphon** to the stems of emergent vegetation. These larvae are found by pulling up aquatic plants (cattails), washing or shaking them in a pan of water (or sampled using a modified bilge pump, Walkers & Crans, 1986), and sieving or searching the bottom muck and debris.

Inspection for *Ae. hendersoni, Ae. sierrensis,* etc., breeding involves searching tree holes and artificial containers such as tires. These containers are often too small for an ordinary dipper, but water can be transferred with a turkey baster or siphoned into a dipper or pan where the larvae can be seen.

ADULT SURVEILLANCE

Adult mosquito surveillance enables evaluation of the incidence of mosquitoes within residential areas where they might bite people and shows the relative abundance of various species. Using this information and reference materials related to breeding sites and habits of the resident species, vector control specialists can determine the need for a control program and conduct an effective search for larval development. Interpreting these observations can provide the justification for applying or withholding control measures.

LANDING AND BITING COLLECTIONS

The required equipment for this method of adult mosquito surveillance is simple and inexpensive. It requires an ethyl-acetate-charged collecting tube or power aspirator, pill boxes, cages for live collections, field record forms or notebook, pencil, flashlight, and map. Although most mosquito control districts use battery-operated mechanical aspirators, a simple mouth-suction aspirator can be made from a section of plastic (or glass) tubing 12 inches long with an inside diameter of about ⅛ inch. Cover one end of the tube with tulle, nylon stocking, or fine wire screening and then insert it into a piece of rubber tubing 2 to 3 feet long. Small pill or salve boxes are convenient for holding dead mosquitoes until they can be identified. A wisp of crumpled soft tissue or lens paper will prevent subsequent damage to the specimens.

Collecting mosquitoes as they bite or land on the surveyor's body is a convenient method of sampling populations. The subject sits quietly for a designated period, usually 5 to 10 minutes, collects the mosquitoes with an aspirator and places them in the collection jar for later identification. It is customary to make landing collections near sundown as this crepuscular period, or twilight period, is the most active time for most mosquitoes. Some individuals are more attractive to mosquitoes than others, so the same person or bait animal might be used throughout a survey. Make collections at about the same time of day and for the same duration, so that activity rates at different stations can be compared to show trends in mosquito abundance. In areas where mosquito-borne disease occurs, landing rates are preferable to biting rates.

Insect sweep net collection

Use insect nets to collect mosquitoes from grass and other vegetation. This type of collection is valuable in determining which species rest in these habitats during the daytime.

Bait trap collection

When other collection methods are inadequate, animal-baited traps, alone or with dry ice, can be used to trap mosquitoes. Make a portable mosquito bait trap from a 12-inch lard can or bucket with inwardly directed screen funnels; bait with young chickens, pheasants, house sparrows, other birds, or about 2 to 3 pounds of dry ice wrapped in newspaper. Commercial traps that convert propane into carbon dioxide, water vapor, and heat to achieve attraction are also available.

Truck trap collection

Some organized mosquito control districts use the basic truck trap, which consists of a large funnel-shaped frame covered with screen and mounted over the roof of a light truck with the entrance (larger) end forward. When driven at low speed (10 to 15 mph) for a fixed distance, mosquitoes caught in the collection bag represent a reasonably unbiased sample in terms of variables in attraction.

Daytime resting collection

Adults of most mosquito species are inactive during the day, resting quietly in dark, cool, humid places. Although labor intensive, careful inspection of daytime shelters can give an indication of the population density of these mosquitoes. This method is especially useful for anopheline mosquitoes. It is also helpful in estimating populations of some culicines, such as *Cx. pipiens*. Because this method does not rely on host-seeking behavior, it is less biased than others, which may be selective for mosquitoes in certain physiological stages. Natural resting stations include houses, stables, chicken houses,

privies, culverts, bridges, caves, hollow trees, and overhanging banks along streams.

A few species of mosquitoes (e.g., *Anopheles* spp.) can be sampled using artificial resting stations such as the red box shelter. This is a wooden box 1 foot wide, long, and deep, with one side open, painted flat black on the outside and red on the inside. Such stations are placed near the suspected breeding places in shaded, humid locations, normally on the ground facing any direction but east. Mosquitoes enter the shelters at dawn, likely in response to changes in light intensity and humidity, and ordinarily do not leave until dusk.

Light trap collection

Light attracts many mosquito species, making it possible to use this response for sampling adult populations between dusk and dawn. Mosquito light traps can attract adults from a considerable distance when placed in locations away from competing light sources. To make the trap even more attractive, place a 2-pound block of dry ice wrapped in newspaper above the trap. The trap should be located 30 or more feet from buildings in open areas near trees and shrubs, and away from other lights, strong winds, and industrial plants giving off smoke or gas. Moonlight may reduce the effectiveness of the traps. The light should be 5.5 to 6 feet above the ground when mounted on a post or hung from a tree. The trap can be operated on a regular schedule from one to seven nights per week; it should be turned on just before dark and turned off after daylight. Remove the collection each morning for sorting and identification. The New Jersey mosquito light trap has been widely used in obtaining data on the density and species composition of mosquito populations. It is powered by 110 volts AC and is placed near a continuous supply of power. The Centers for Disease Control (CDC) battery-operated aspirators or vacuum collectors can be used for the same function as light traps. Many mosquito control districts rely on these traps to provide a comparable count with historical data.

The light trap was developed as a portable option for collecting live mosquitoes in areas where standard electric power is not available. As the mosquitoes respond to the attractants, they are blown downward through a screen funnel into a killing jar or a mesh bag suspended below the trap. The light trap collects a high percentage of mosquitoes in proportion to other insects and many more females than male specimens, a desirable feature in collecting mosquitoes for virus studies.

Different species of mosquitoes have considerable differences in their reactions to light. Light trap collections are often used to complement other methods of sampling mosquito populations. These traps can be converted to monitor densities of mosquitoes not attracted to light by removing the light and using only carbon dioxide as the attractant. With light as an attractant, these traps have proven very useful in measuring densities of some of the culicine mosquitoes, such as *Ae. vexans* and *Cq. perturbans*. Some anophelines, especially *An. walkeri,* are also readily collected in light traps. The use of carbon dioxide only as an attractant collects large numbers of *Ae. canadensis, Ae. fitchii, Ae. idahoensis,* and *Ae. intrudens*. However, members of the *Cx. pipiens* species complex and *Ae. aegypti* are seldom taken in significant numbers. Light trap collections may fluctuate with the dark and bright phases of the moon (collections are greatest during the dark phases). The effect of moonlight can be offset to some extent by placing traps in locations shaded from the moon.

Gravid trap collection

This trap has proven to be very effective in capturing large numbers of egg-filled (**gravid**) female *Culex* mosquitoes. Gravid traps are easy to build, lightweight, portable, and run on one 6-volt gel cell rechargeable battery. One gallon of attractant (infusions made from grass, rabbit pellets, manure, etc.) placed in a black tray draws gravid females to the trap to oviposit. While flying above the attractant's surface, the mosquitoes are drawn into the collection chamber by an air current created by a motorized fan. The mosquitoes are captured live and then can be used for virus-isolation studies or species density information.

Other trap collection

The BG Sentinel trap is a relatively new trap that has quickly become the gold standard for collecting the introduced container-breeding *Ae. aegypti* and *Ae. albopictus* mosquitoes. It incorporates a design meant to mimic human heat convection with a specially developed lure that has three human-emitted attractants. When coupled with CO_2, this trap enables mosquito control professionals to effectively sample the usual suspects with the option for detecting introductions of these new (to Wyoming) exotic species.

USING MOSQUITO SURVEY DATA

Data from the preliminary reconnaissance surveys are correlated with reported disease prevalence or complaints of pest mosquitoes. But it is only after reviewing all the information that the health office or mosquito control supervisor can make an intelligent decision as to the need for a control program and the type of control operations that will be most effective and economical.

Inspections that have been continued routinely once a mosquito control project is under way are used to evaluate progress. Success or failure of a mosquito control project cannot be measured in terms of the number of feet of ditches constructed or the number of gallons of insecticides used. While these statistics may be useful for some purposes, it is the actual density of mosquito populations that is significant. If the density is reduced to a satisfactory level, the routine surveys will reflect this reduction and document the accomplishment. On the other hand, if mosquito populations remain high, these results will spur intensified efforts to obtain the desired level of control. Additionally, for comparison, it is advisable to inspect some comparable untreated breeding areas at regular intervals to determine the normal fluctuation of various species throughout the season as well as between seasons.

The correlation between mosquito annoyance and numbers captured in light traps has been established in many localities. In one state, for example, it was determined that general annoyance did not ordinarily occur until the number of female mosquitoes of all species exceeded 24 per trap per night. Similar criteria can be worked out for other areas and for various species, taking care to note that the nuisance capacity of species varies greatly. Meaningful action thresholds can provide justification for application decisions. Counts of mosquito larvae are a bit more difficult to correlate with pest problems or disease hazards. However, larval surveys reveal the specific sources of mosquito production. This information is invaluable to the control supervisor, who can then apply effective **larvicides** to the right places at the right times to keep adult mosquito populations below disease-vectoring or annoyance levels. Data over a period may also serve to justify the use of permanent control measures, such as source reduction. Expensive operations, such as filling and draining, should be undertaken only when careful inspection of each area has shown its role in the production of the vector or pest species that is important in the locality.

ARBOVIRUS SURVEILLANCE IN VECTORS AND HOSTS

Mosquitoes have a long history of spreading disease. Despite modern medical advances and various control efforts, the World Health Organization cites hundreds of millions of cases of malaria, dengue, and yellow fever afflicting tropical and subtropical residents annually. Add rapidly spreading emerging diseases such as Rift Valley fever in domesticated animals and Chikungunya and Zika virus in humans, and mosquitoes' impact spreads exponentially. Effectiveness of mosquito control programs where mosquito-borne diseases are a concern clearly depends upon early detection of disease and prompt, skilled response. Enzyme-linked immunosorbent assay (ELISA) and polymerase chain reaction (PCR) methodology are lab-based methods for testing reservoir blood samples for West Nile virus (WNV), Saint Louis encephalitis (SLE), Western equine encephalitis (EEE), and other **arboviruses** (**ar**thropod-**bo**rne **viruses**). These tests yield results quickly. Recently, district-level technology in the form of Response Biomedical's rapid analyte measurement system (RAMP) allows mosquito control personnel to test non-blood products for virus detection. Surveillance methods leading to such tests include frequent communication with other mosquito control workers, healthcare workers, and veterinarians; collection of moribund wild birds or animals; use of sentinel flocks of chickens, quail, or pheasants; and testing pooled mosquito samples of the species most likely to obtain and transmit viruses.

COMMUNICATING WITH OTHER PROFESSIONALS

State arbovirus surveillance committees maintain frequent contact with public health and veterinary laboratories to collect and report information on mosquito-borne arbovirus activity. This information is compiled by the Wyoming Department of Health and shared with interested mosquito control personnel in the form of a weekly West Nile virus update with human, equine, avian, and mosquito surveillance data. Weekly updates and information on how to sign up to receive the emails is available on the Wyoming Mosquito Management Association website (https://sites.google.com/view/wyomosquito). In addition, mosquito control personnel keep in touch with local healthcare personnel and veterinarians about indications of arbovirus activity in the area. If cases are suspected, that information is passed along. It is important to note that not all cases of encephalitis are mosquito borne. Caution must be exercised in reacting to initial reports, and appropriate agencies need to work closely to get laboratory confirmation of suspected cases. In some less critical cases of disease, some healthcare practitioners are reluctant to subject patients to confirmatory testing because confirmation of the cause of the condition (encephalitis, meningitis, or poliomyelitis) would not alter the treatment protocol. It is imperative to communicate the need for confirmatory testing to local healthcare practitioners for the sake of providing surveillance data that mosquito control personnel may act on to prevent further disease cases.

SAMPLING WILD BIRD POPULATIONS

As active handling of live wild birds requires a permit, it is not likely that mosquito personnel in Wyoming, unless cooperating with the Wyoming Game and Fish Department, would have access to sample sickly birds. However, passive surveillance for West Nile virus morbidity or mortality in either reservoir (American robins, house sparrows, house finches, doves) or indicator species (crows, ravens, jays, and magpies) allows mosquito abatement personnel to detect hot spots of viral activity and react accordingly, generally by saturating an area with mosquito traps to detect virus therein.

SENTINEL FLOCKS

Chickens, quail, pheasant, or other sentinel birds are retained in outdoor cages in specific sampling areas and bled periodically to monitor arbovirus activity. These sentinel birds are raised in a mosquito-free environment and tested prior to placement to ensure that they have not been exposed to arboviral activity elsewhere. If the sentinel bird tests positive after being placed in an area, it is a sure sign of arbovirus activity in the area. A supply of unexposed birds should be readily available to replace those that become infected. To adequately sample large areas requires numerous sentinel flocks, so this method can be costly.

ARBOVIRUS SURVEILLANCE IN MOSQUITOES

Perhaps the best early warning method available is the isolation of viruses from wild mosquitoes. Because female mosquitoes must feed on an infected animal before they can pick up a virus, collect female mosquitoes that have had at least one blood meal as indicated by an abdomen engorged with fresh or partially digested blood meals, or containing eggs. There are several sampling methods and procedures that will help increase the ratio of engorged and gravid *Culex*, but this is more difficult with *Aedes, Anopheles, Culiseta, Coquillettidia, Ochlerotatus,* or *Psorophora*. Aspirate *Culex* from dark closets, cabinets, or other areas of abandoned homes and vacant buildings whose open windows or doors are attractive to *Culex* mosquitoes seeking places to rest during the day. If the buildings are sampled on a regular basis, the mosquitoes collected probably will have fed on a variety of hosts. Gravid traps are also useful for collecting gravid specimens of adult *Culex* as well.

Light traps and carbon-dioxide-enhanced traps can be used effectively in wooded areas, storm drains, catch basins, manholes, tire piles, etc., but they tend to collect many mosquitoes that have not had a blood meal. Resting boxes can be used to collect some important vector species (e.g., **anophelines**), but they do not necessarily bias the collection toward gravid or engorged females, and they do not normally collect large numbers of mosquitoes.

TRANSPORTING MOSQUITOES

Store live mosquitoes in a cool, moist container, and identify and pool them as soon as possible. An ice chest containing a small amount of ice (not dry ice) is ideal for transporting mosquitoes, taking care that the mosquitoes do not get wet during shipment.

HOLDING, IDENTIFYING, AND POOLING COLLECTIONS

Once in the laboratory, mosquito collections can be frozen and should not be left outside the freezer for extended periods of time during identification. Mosquitoes from each collection are identified separately and a fixed number, usually 50 or fewer of the same species collected at the same location and time, will be tested for viruses. Most mosquito control entities in Wyoming use the Response Biomedical rapid analyte measurement platform (RAMP) system for West Nile virus detection. This system uses a special camera to detect fluorescent markers attached to antibodies that adhere to a specific portion of the virus if it is present. Use of a chill table to identify, sort, and pool adult mosquito samples helps keep specimens cool and preserves viruses that may be present in them.

CHAPTER 4: CONTROL

LEARNING OBJECTIVES

1. **Understand the principles of integrated pest management (IPM).**
   - Define integrated pest management (IPM) and explain its significance in mosquito control.
   - Identify the components of IPM used in mosquito control programs, including source reduction, habitat modification, biocontrol, and pesticide applications.
2. **Evaluate mosquito control strategies.**
   - Compare the effectiveness of targeting mosquito larvae versus adult mosquitoes.
   - Analyze the environmental and public health implications of using larvicides and adulticides.
3. **Implement source reduction techniques.**
   - Describe various source reduction methods, from simple actions like overturning containers to complex water management practices.
   - Explain how effective source reduction can minimize or eliminate the need for chemical interventions.
4. **Apply water management for mosquito control.**
   - Discuss water management strategies to prevent mosquito breeding, including manipulation of water levels and open marsh water management.
   - Recognize the regulatory requirements for water management projects, particularly in Wyoming.
5. **Utilize biological control methods.**
   - Identify common biological control agents used in mosquito control, such as larvivorous fish.
   - Evaluate the benefits and limitations of biological control methods in mosquito management.
6. **Select and apply larvicides.**
   - Differentiate between various larvicides and their modes of action, including bacterial toxins, insect growth regulators, and conventional insecticides.
   - Determine the appropriate larvicide formulation and application method based on habitat characteristics and mosquito species.
7. **Operate and calibrate larvicide application equipment.**
   - Describe the equipment used for larvicide applications, including ground and aerial equipment.
   - Perform calibration of larvicide application equipment to ensure accurate dosing.
8. **Conduct adulticide applications.**
   - Explain the role of adulticide applications in mosquito control and when they are considered necessary.
   - Discuss the importance of droplet size and application timing in maximizing adulticide efficacy and minimizing environmental impact.
9. **Implement barrier treatments and space sprays.**
   - Describe the use of barrier treatments and space sprays for localized mosquito control.
   - Understand the precautions necessary to protect non-target organisms and ensure human and pet safety.
10. **Manage insecticide resistance.**
    - Explain the importance of resistance management in mosquito control programs.
    - Outline strategies to prevent and manage insecticide resistance, including rotating chemicals and conducting regular resistance testing.
11. **Explore genetic manipulation techniques.**
    - Discuss advanced mosquito control methods involving genetic manipulation, such as the use of *Wolbachia* bacteria and genetically modified mosquitoes.
    - Evaluate the potential benefits and ethical considerations of these emerging technologies.

While it is not possible to provide a concise, generic overview of all mosquito control programs in the U.S., there are certain components that virtually all operational programs include as they are inherent to the principles and practices of integrated pest management (IPM). Traditionally an overriding concept in organized mosquito control, the IPM acronym was not used in this sense a century ago when mosquito control was in its infancy. Typical IPM programs use a combination of resource-management techniques that include source reduction, habitat modification, biocontrol, and larvicide and adulticide applications, all based on surveillance data as to need and timing of application. Continuing education, for both employees and the public, is also an important component of most mosquito control programs. This has resulted in a more informed public and increased professionalism among mosquito control workers, where significant progress has been made in attempting to reduce pesticide use and risk.

An important consideration in the practice of mosquito control is the advisability, whenever possible, to target control operations against larvae. They are usually concentrated and relatively immobile and therefore occupy minimum acreage compared to adults, which may rapidly spread over large areas. By targeting the larvae, it is possible to minimize the area treated and often avoid treating areas with high human activity. Conversely, targeting adult mosquitoes may require highly visible and extensive applications of **adulticides** within residential and urban areas. The adulticides registered for this use are applied at levels 100 to 10,000 times below rates that would be cause for concern about exposure risk for the public or the environment. Nevertheless, achieving good larval control while at the same time minimizing the use of adulticides is environmentally and client friendly, and is appreciated by the public. The use of pesticides for mosquito control is considered a temporary form of control. Because it is unreasonable to expect to eliminate the next generation by treating the current generation, the process may have to be repeated time and again. This is true for chemical treatment of both larval and adult populations of mosquitoes. The need for this type of temporary control can be reduced by implementing permanent control measures whenever and wherever possible. Permanent control measures are discussed in the source reduction section. These methods include land and water management approaches that prevent breeding. Where successful, they can be permanent in nature, perhaps requiring limited maintenance from time to time. Although this activity may escape the attention of the public, it is extremely effective in easing the management burden of protecting the public from mosquitoes.

SOURCE REDUCTION

Source reduction ranges from the simple overturning of a discarded bucket or disposing of waste tires to complex water-level manipulations in wetlands. The removal or reduction of mosquito breeding habitat is often the most effective and economical long-term method of mosquito control. These efforts can often minimize, or at best eliminate, the need for mosquito larvicide in the affected habitat and can greatly reduce applications of adulticide in nearby areas.

SANITATION

Containers of all types, and man-made structures such as horse troughs, roof gutters, and discarded tires, can produce high numbers of mosquitoes, including species that can transmit several **pathogens**. Removal of debris and regular inspection can reduce breeding in such sites. Typically, mosquito control-related sanitation efforts are best accomplished by homeowners and

residents who have inadvertently created mosquito breeding sites around their homes (known as **peridomestic habitat**). Mosquito control agencies often support educational programs that call attention to the hazards and assist individual efforts in residential area cleanup.

WATER MANAGEMENT

Prevention of breeding can be accomplished by removing surface water from productive sites to reduce **oviposition**. This involves frequently manipulating water levels or flushing areas in which mosquitoes are developing. On ranches, these water manipulation approaches can include irrigating to only sub-surface saturation of soils, flooding hayfields and pastures with short pulses of water that do not offer sufficient time to complete development to pupae, and/or rapid flushing of the immatures into rivers, where they are unlikely to survive.

Although originating in coastal salt marsh habitats, the practice of **open marsh water management** (OMWM) can also be applied in freshwater scenarios in Wyoming. This is a technique in which mosquito-producing locations on the marsh surface are connected to deep water habitat (e.g., tidal creeks, ponding areas, deep ditches, etc.) with shallow ditches. Mosquito broods are thus controlled without pesticide use by providing access for naturally occurring larvivorous fish to the mosquito-producing depressions or, conversely, by naturally draining these locations before adult mosquitoes can emerge. These hydrological connections between marsh and estuary can enhance natural resources, such as waterfowl populations and fisheries, and benefit mosquito control. The use of shallow ditching (approximately 3 feet or less in depth) rather than the deep ditching used in years past is considered more environmentally acceptable because with shallow ditches, fewer unnatural hydrological impacts occur to the marsh. In Wyoming, maintenance of existing man-made ditches falls under the jurisdiction of the Wyoming Department of Environmental Quality, whereas new projects likely fall under the permitting purview of the U.S. Army Corps of Engineers. In all situations, refer to the appropriate regulatory agencies prior to planning any work in wetland areas.

BIOLOGICAL CONTROL

Biocontrol is the use of biological organisms to control pests, in this case, insect pests. Biocontrol is popular in theory because of its potential to be host specific with virtually no **nontarget** effects.

Overall, larvivorous fish are the most extensively used biocontrol agent for mosquito control. Predaceous fish, such as *Gambusia affinis* and other top-feeding minnows (Poeciliidae) and killifish (Cyprinodontidae) that occur naturally in many aquatic habitats, can be collected (or in some cases propagated) and placed in permanent water bodies for larval control. In Wyoming, consultation (and possibly permitting) with state Game and Fish officials is critical prior to any collection, relocation, or introduction of fish for mosquito control purposes. This is necessary to eliminate any unintended impacts to native fish via competition, predation, or disease.

Other biocontrol agents have been tested for use as mosquito control but to date have not generally been operationally feasible. These include the predaceous mosquitoes of the genus *Toxorhynchites,* predaceous copepods, the parasitic nematode *Romanomermis,* and the fungus *Laegenidium giganteum*. Biocontrol certainly holds the possibility of becoming a more important tool and playing a larger role in mosquito control in the future when improved technology and more attractive economics may enhance its usefulness.

Unfortunately, there is no basis for the claimed suppression of mosquito-biting activity, due to the feeding of bats or purple martins. While much of their often-touted voracity for mosquitoes was derived from simulated studies in laboratories, their predation is insufficient to significantly impact mosquito density in the wild.

LARVICIDAL APPLICATIONS

The application of pesticides to kill immature mosquitoes by ground or aerial applications is typically more effective and target specific than the application of an adulticide, yet less permanent than source reduction. Several pesticides in various formulations are labeled for mosquito larval control. These include **biorational** products with active ingredients derived from various bacteria such as *Bacillus thuringiensis* var. *israelensis* (**Bti**), *Bacillus sphaericus* (**Bs**), and *Saccharopolyspora spinosa* (spinosad), as well as insect growth regulators (**IGRs**), including juvenile growth hormone (methoprene) and chitin synthesis inhibitors (novaluron). Also labeled for mosquito control are conventional insecticides, including organophosphate, temephos, several mineral-based or plant-derived oils, and monomolecular films. The choice of larvicide and, more specifically, the formulation, is driven by the species present, the habitat of the targeted larvae (including factors such as hydrology and vegetation), the persistence of the larval recruitment, and the presence of desirable nontarget organisms.

The timing of **larvicide** application is dependent on the nature of the control agent. Conventional insecticides, for example, kill **larvae** at all stages and thus can be applied when convenient. Bacterial toxins of Bti and Bs must be consumed by the larvae and are usually applied well before the fourth instar to ensure that consumption occurs. The juvenile hormone class of IGRs mimics an essential hormone that is present in high concentration in early instar larvae but in very low concentration in late (fourth) instar larvae. Exposure of fourth instar larvae to the hormone upsets the physiological molting process and kills mosquitoes in the subsequent pupal stage.

IGRs can be formulated as slow-release insecticides so that application in the second or third instar will result in an adequate exposure during the fourth instar. Chitin synthesis inhibitors affect the ability of the larvae to reattach their muscles to the **exoskeleton** during the molting process and thus are effective throughout the entire larval life. Monomolecular films prevent the insect from remaining at the surface of the water by reducing surface tension. Under these conditions, larvae and pupae deplete their energy reserves trying to stay at the surface and succumb to exhaustion. Oils (from all sources) kill larvae and pupae by suffocation because the insects are not able to obtain air through their **siphon** at the oily surface.

Thus, each larvicide has very specific applications and may be more effective against one species or group than another. Each label is different, and special attention must be given to a full understanding of the provisions of the label for each chemical being considered for use in mosquito control programs. The label prescribes application methods and rates, habitat restrictions, personal and environmental exposure limits, etc. Pesticide applicators must be knowledgeable about the label contents and abide by the label.

Important factors for larvicide choice include specificity for mosquitoes, minimal impact on nontarget organisms, and, in many instances, the ability to penetrate dense vegetative canopy. Larvicide formulations (e.g., liquid, granule, pellet, water-dispersible granule, water-soluble pouch, briquet) must be accurately applied and appropriate to the habitat being treated.

Larvicidal applications, when based on accurate surveillance data, are an integral component of an integrated mosquito control operation. Finally, accuracy of application coverage is important, as failure to cover even a relatively small portion of a breeding area can result in the emergence of a large mosquito brood and lead to the need for immediate broad-scale applications of adulticide.

LARVICIDE APPLICATION EQUIPMENT

Larvicide treatments can be applied with either ground or aerial equipment. Ground applications are dependent on the type of formulation being applied. Dry formulations, such as granular and pelleted larvicides, are typically applied with power blowers or crank-operated spreaders whereas briquette or dunk formulations are generally applied by hand. Liquid formulations, such as emulsifiable concentrates, liquid concentrates, or wettable powders, are applied with hand sprayers or field sprayers. Choose formulations based on accessibility to the area, size of the treatment area, amount of aquatic vegetation present, residual activity, and equipment available.

Always calibrate your equipment prior to application to ensure that the correct dose is obtained; refer to the Wyoming Pesticide Applicator Certification Core Manual for calibration of hand sprayers and boom sprayers. Even power blowers and crank spreaders can be calibrated. Calibration of power blowers and hand-crank spreaders is essentially a very simple process:

Spreader calibration

1. Measure the width of spread.
2. Apply the granules to a small area.
3. Measure the amount applied to this area.
4. Compare this amount to the desired dose and adjust the spreader settings until the desired output is obtained.

It is important to note that calibration of hand-crank spreaders is dependent on consistent speed of application and the consistent turning of the hand crank (generally one rotation per second).

Power blower calibration

1. Weigh the amount of product before application.
2. Apply the granules to a small area.
3. Weigh the amount of product after application and determine the amount applied to this area.
4. Compare this amount to the desired dose and adjust the blower's settings until the desired output is obtained.

Aerial equipment to apply larvicides is costly and generally justified for treatment areas too large for or inaccessible by ground equipment, or only under emergency conditions. These applications can be made by airplanes, helicopters, and unmanned aerial vehicles (UAVS, also known as drones). Since applications are made above the target area, aerial applications are greatly affected by wind speed, direction, and canopy density. Make sure to use products that are dense enough to minimize the effect of wind and rotor wash from helicopters. Poorly maintained equipment (airplane, helicopter, or UVA) affects the application accuracy and rate. Aerial equipment requires precise calibration.

ADULTICIDE APPLICATIONS

The ground or aerial application of insecticides to kill adult mosquitoes is usually the least efficient mosquito control technique and is considered the last resort. This option is reserved for managing mosquito populations that have reached the adult stage despite efforts to intervene in the larval stage or when such treatment has not been conducted. The tendency of poorly funded or misguided mosquito control organizations to use only adulticides and bypass the other, often more effective, options available conflicts directly with accepted practice. Nevertheless, applications of adulticides, when based on concurrent surveillance data, is an extremely important part of the **IMM** (integrated mosquito management) approach when undertaken with the appropriate amount of insecticide specified on the label.

Adulticide application equipment

Adulticides are often applied as ultra-low volume (ULV) sprays in which small amounts (3 fluid ounces or less per acre) of insecticide are dispersed either by truck-mounted equipment, or aerially from airplanes, helicopters, or unmanned aerial vehicles (UAVs). Ground or aerially applied thermal (fogging) application of adulticides is still used in some areas but to a much lesser degree. Mosquito adulticide ULV application differs fundamentally from efforts to control many other adult insects.

The fine ULV droplets (high concentration but very low dosage) must drift through the habitat and directly contact flying mosquitoes to provide optimal control benefits. These applications are scheduled to occur between dusk and shortly after dawn, when most beneficial insects are resting. These tiny droplets are unlikely to affect insects larger than mosquitoes because those larger insects' physical characteristics and size cause the droplets to drift around rather than deposit on them. The very small flying mosquitoes, however, do not ward off the small droplets in this manner and are susceptible to the lethal dose held within those small droplets. Also, the fine ULV droplets means that there are literally millions more droplets to directly contact mosquitoes than there would be if larger droplets were used.

Achieving and maintaining appropriate droplet size and dispensing rate should not be left to chance. Calibration and droplet testing of all ULV sprayers are required to occur annually or whenever a new product is used, or when the machine is serviced. Specialized equipment is required to calibrate and conduct droplet testing on these sprayers. Fortunately, support with the technology required to perform such testing and calibration is readily available from several of the companies that sell mosquito control products in Wyoming. After calibration, be sure to monitor the mix ratio (active ingredient to diluent), application speed, air temperature, wind speed, pressure, generator engine speed, flow rate, and droplet size throughout the application to ensure the labeled rate is applied. In addition, it is important to ensure proper working order of nozzles, lines, and fittings.

Using the proper size range for the droplets makes it possible to increase control efficiency and decrease the risk of adverse impact to the environment and public health. The small droplets drift far beyond the point of release and settle in a widely dispersed manner on the ground, often after their toxicity has been degraded by **hydrolysis** (chemical breakdown due to moisture). The relatively minuscule amount of toxicant in each droplet further protects against adverse impact. Thus, while the technique lends itself to the criticism that nontarget organisms can be impacted, adherence to the label specifications for droplet size in ground applications confines the possibility of adverse impact to relatively few, small nocturnal organisms. This is a constant consideration for control programs, especially those relying heavily on aerial applications of adulticides, for which the droplet size spectrum has traditionally been somewhat larger. Recent studies (Lesser, 2017) have demonstrated that newer aerial rotary spray nozzles can achieve that smaller droplet size in the air and can offer a concomitant decrease in the flow rate necessary for effective control. Extended experience, coupled with EPA studies, demonstrates that when applied according to the label (the law), these applications have minimal or no effect on most nontarget organisms.

BARRIER TREATMENTS

Barrier treatments are typically applied as high volume (low concentration) liquids with conventional applications like a mist blower and hand-held spray equipment using compounds with residual characteristics. This method is common in some U.S. locations and its use is growing. This technique is especially attractive to individual homeowners living near mosquito-producing habitats where residual chemicals applied to the vegetation along property borders can provide relief to the residents, occasionally the only recourse

available to battle mosquitoes arriving from non-treated areas. This treatment is also being used for the perimeter of small harborage areas, such as un-mowed grass and vegetation under bleachers, associated with parks and recreation areas, walking/biking trails, or areas around public events. This practice includes using attractive toxic sugar baits (ATSB), which are applied to vegetation or in stations intended to lure mosquitoes to feed and ingest a lethal dose of a toxin-laced sugar solution. Care must be taken to avoid applying to flowering plants with pollinator activity as the active ingredient is not always mosquito specific.

SPACE SPRAYS

Space sprays are generated by portable ULV equipment, and are typically used to provide indoor mosquito control in houses, tents, trailers, warehouses, etc. For small enclosures, commercial aerosol (bug bomb) applications are also highly effective. These applications require close review of the label to ensure the safety of inhabitants and their pets when they re-enter the establishment after completion of the application. This technique relies on the movement of fine droplets throughout the enclosed space to directly contact the mosquitoes. Alternatively, in certain circumstances residual applications of insecticides are placed on interior walls to kill mosquitoes that subsequently rest on the treated surfaces. Residual treatments, common overseas, are not routinely used in the U.S. for mosquito control, but some insecticides are labeled for use. Adulticides labeled for mosquito control include organophosphates, natural pyrethrin, and synthetic pyrethroids. Studies are underway to incorporate spatial repellent spray use of plant terpenes such as oil of citronella (and 150+ other such plant extracts) to synergize pyrethroid adulticide efficacy in areas with demonstrated resistance (Norris et al., 2015). As with other pesticides, the specific attributes and methods of use for adulticides are listed on the label. It is incumbent on the pesticide applicator to apply the product as directed by the label.

RESISTANCE MANAGEMENT

Insecticide resistance testing is a surveillance tool that determines if a population of mosquitoes is developing a tolerance to a particular active ingredient. This tolerance may result from natural genetic variability, increased metabolic detoxification of the active ingredient, or a behavioral change that shields the mosquitoes from exposure. Awareness of the susceptibility of local mosquitoes is of extreme importance to an adulticide program because the variety of available pesticide classes (pyrethroids and organophosphates) is very limited. An agent with compromised efficacy is one less tool available when a disease outbreak threatens lives. Insecticide resistance is commonly tested using the **bottle bioassay resistance test** and is described and demonstrated at https://www.cdc.gov/mosquitoes/php/toolkit/cdc-bottle-bioassay.html. Some methods to limit the development of resistant populations include using larvicides and adulticides with varying modes of action/active ingredients; preemptively rotating the classes of adulticides or larvicides used; and ensuring that a lethal dose is applied each time. This last point hinges on maintaining accurate and frequent (at least annual) calibration and droplet testing of all ULV spray equipment and applying products at appropriate label rates.

GENETIC MANIPULATION OF MOSQUITOES

Beyond widely used adult control methods, there are several alternative strategies. These include endoparasitic approaches using *Wolbachia* bacteria, sterile insect technique, and most recently, genetically modified mosquitoes developed to pass along a dominant lethal gene to offspring. *Wolbachia* is a bacterium that infects many different species of insects in nature. When introduced into uninfected mosquito populations, its presence can greatly reduce reproductive success through a process of cytoplasmic incompatibility. Infected males pass the bacteria to the mated female's subsequent egg batches, making them unviable. Alternately, females carrying the bacterium create viable offspring, also carrying *Wolbachia*, that spread it to future generations. Additionally, *Wolbachia* infection has been shown to prevent mosquito acquisition of several pathogens including dengue, Chikungunya, and yellow fever viruses, thus making the populations of *Wolbachia* carriers unlikely to obtain and transmit certain diseases. Closely related sterile insect technique (SIT) and RIDL, release of insects containing a dominant lethal (gene), introduce lab-reared and modified adults to wild populations that pass along sterility to offspring or a dominant gene that causes mortality. While these technologies may seem based on science fiction of the recent past, rapidly advancing genetic techniques are making these approaches to mosquito population and disease control very much available.

CHAPTER 5: PERSONAL PROTECTION AND EDUCATION

LEARNING OBJECTIVES

1. **Understand personal protection measures.**
   - Identify the recommended personal behavior practices to minimize mosquito bites and reduce the risk of mosquito-borne diseases.
   - Recognize the importance of wearing appropriate clothing and using repellents such as DEET, picaridin, IR3535, and oil of lemon eucalyptus, according to CDC guidelines.
2. **Learn environmental management techniques.**
   - Describe methods for reducing mosquito breeding sites around homes, such as removing water-holding containers and cleaning roof gutters.
   - Understand the impact of proper housing and environmental management in minimizing mosquito exposure.
3. **Appreciate the role of education and information sharing.**
   - Explain the significance of continuous education for mosquito control professionals to stay updated with the latest research and technologies.
   - Identify key resources and organizations that provide information and support for mosquito control, such as the American Mosquito Control Association, CDC, and local health departments.

The public and the pesticide applicator must consciously adopt personal behavior practices that reduce the probability of receiving infective mosquito bites. For example, most, but not all, vectors are quite active during sunset and sunrise. This is a time of day when exposure to mosquitoes should be minimized. Staying indoors as much as possible, especially if there is an alert situation in connection with mosquito-borne disease, is one way to avoid contact. If it is necessary to go outside, wear light-colored clothing (which is less attractive to mosquitoes), pants and long sleeves, and apply a repellent to the exposed skin—carefully following the manufacturer's directions on the label.

The Centers for Disease Control and Prevention (CDC) recommends three other active ingredients for repellents in addition to the gold standard of DEET. These include picaridin, IR3535, and oil of lemon eucalyptus (PMD). For more information on recommended repellents and their proper use, visit https://www.cdc.gov/mosquitoes/prevention. Remove water-holding containers from the yard, clean roof gutters, and use other physical means to reduce breeding near the home. Avoid known mosquito habitats, if possible, and keep personal exposure to a minimum.

In the U.S., residents are fortunate that mosquito-borne disease transmission to humans is relatively rare, and infected people often do not have overt symptoms. Theoretically, thousands of infections could occur annually without causing clinical symptoms, but in a low percentage of cases infection can lead to disease and even death. So, prudence is the watchword. Personal protection is a means to reduce the probability of infection. There are other reasons for the relatively low incidence of human involvement with mosquito-borne diseases in the U.S., but we often overlook the obvious by building homes at the margins of wetlands, thereby multiplying the probability of mosquito encounter and infection. However, organized mosquito control, affordable housing, proper screening, indoor plumbing, air conditioning, and television all contribute to reducing human exposure to mosquitoes, even in high-mosquito-density areas. Societies in tropical and subtropical climates are sometimes less fortunate in terms of the physical protection from mosquitoes that is afforded by their surroundings.

Mosquito control professionals are the locally recognized experts on the topics of mosquitoes, their control, and often, their associated diseases. Staying current with the latest mosquito research and technologies and sharing interpretations of that information with the local public is an integral part of a true integrated mosquito management program. Because mosquito-borne diseases and the control response necessary to curtail disease outbreaks often elicit strong responses from the public, having information readily available to share with local residents and stakeholders is critical *before a crisis arises.*

Being informed and prepared cannot be overstated. Resources for such continuing education include the website www.mosquito.org, frequent webinars, and annual meeting of the American Mosquito Control Association; the Centers for Disease Control and Prevention Division of Vector-Borne Diseases (https://www.cdc.gov/vector-borne-diseases); the Wyoming Department of Health Infectious Disease Epidemiology Unit (https://health.wyo.gov/publichealth/infectious-disease-epidemiology-unit/disease/west-nile-virus) and the website, annual meeting, and support of your fellow mosquito control professionals and product representatives at the Wyoming Mosquito Management Association (https://sites.google.com/view/wyomosquito).

GLOSSARY

Adulticide: A type of pesticide used to kill adult mosquitoes.

American Mosquito Control Association (AMCA): A professional organization that provides resources, education, and support for mosquito control professionals.

Bottle bioassay resistance test: A surveillance tool used to determine if mosquito populations are developing resistance to insecticides.

Centers for Disease Control and Prevention (CDC): A U.S. federal agency that provides guidelines and information on public health issues, including mosquito-borne diseases and recommended repellents.

Crepuscular periods: Times of day around dawn and dusk when many mosquito species are most active.

DEET: A common active ingredient in insect repellents, known for its effectiveness in preventing mosquito bites.

Educational programs: Initiatives aimed at informing the public about mosquito control practices and the importance of reducing mosquito breeding sites.

Epidemiology: The branch of medicine that deals with the incidence, distribution, and control of diseases, including those spread by mosquitoes.

Integrated mosquito management (IMM): A comprehensive approach to mosquito control that includes surveillance, source reduction, habitat modification, biological control, and the use of larvicides and adulticides.

IR3535: Another CDC-recommended active ingredient in insect repellents, known for its effectiveness and safety.

Larvicide: A type of pesticide specifically designed to target and kill mosquito larvae before they mature into adults.

Mosquito control professionals: Experts trained in the methods and practices of mosquito control, including surveillance, pesticide application, and public education.

Oil of lemon eucalyptus (PMD): A plant-based active ingredient in insect repellents, recommended by the CDC as an alternative to DEET.

Picaridin: An active ingredient in insect repellents, recommended by the CDC as an alternative to DEET.

Peridomestic habitat: Areas around human dwellings where mosquitoes can breed, such as gardens, yards, and patios.

Personal protection: Measures taken by individuals to prevent mosquito bites, such as using repellents and wearing protective clothing.

Source reduction: The elimination or modification of mosquito breeding sites to reduce the mosquito population.

Sterile insect technique (SIT): A method of pest control that involves releasing sterilized males to reduce the population by mating with wild females, resulting in no offspring.

Ultra-low volume (ULV) sprays: A method of applying insecticides in very small amounts to create a fine mist that can effectively control adult mosquito populations.

Wolbachia: A type of bacteria used in mosquito control that reduces the ability of mosquitoes to reproduce and transmit diseases.

REFERENCES

Myers, C.M. (1967). Identification and description of *Aedes* eggs from California and Nevada (Diptera: Culicidae). *Canadian Entomologist*, 99, 795–806.

Bosak, P. J. and W. J. Crans. 2002. The structure and function of the larval siphon and spiracular appa ratus of *Coquillettidia perturbans. J Am Mosq Control Assoc*. 18(4):280-283.

Carpenter, S. J. and W. J. LaCasse. 1955. Mosquitoes of North America (North of Mexico). University of California Press, Berkeley, CA.

Clements, A. N. 2008. The biology of mosquitoes: development, nutrition, and reproduction. CABI Publishing, Oxfordshire, UK.

Darsie, R. F. Jr. and R. A. Ward. 2005. Identification and geographical distribution of the mosquitoes of North America, North of Mexico. University of Florida Press, Gainesville, FL.

Edmund J. N., A. D. Gross, B. M. Dunphy, S. Bessette, L. Bartholomay and J. R. Coats. 2015. Comparison of the Insecticidal Characteristics of Commercially Available Plant Essential Oils Against *Aedes aegypti* and *Anopheles gambiae* (Diptera: Culicidae). *J Med Entomol*. 52 (5): 993-1002. doi: 10.1093/ jme/tjv090

Huang, S., D. J. Smith, G. Molaei, T. G. Andreadis, S. E. Larsen and E. F. Lucchesi. 2013. Prevalence of *Dirofilaria immitis* (Spirurida: Onchocercidae) infection in *Aedes, Culex, and Culiseta* mosquitoes from north San Joaquin Valley, CA. *J Med Entomol*. 50 (6): 1315-1323.

Knight, D. H., G. P. Jones, W. A. Reiners, and W. H. Romme. 2014. Mountains and plains: The ecology of Wyoming landscapes, 2nd ed. Yale University Press, New Haven, CT.

Lesser, C. Efficacy of aerially applied Fyfanon (malathion) at rates below EPA-label prescription. Presented 2/14/17. American Mosquito Control Association Annual Meeting, San Diego, CA.

Lunt, S. R. and G. E. Peters. 1976. Distribution and ecology of tree-hole mosquitoes along the Missouri and Platte Rivers in Iowa, Nebraska, Colorado, and Wyoming. *Mosquito News*. vol. 36(1): 80-84.

O'Malley, C. 1995. Seven ways to a successful dipping career. *Wing Beats*. vol. 6(4): 23-24. http://vectorbio.rutgers.edu/outreach/dipping.htm

Rees, D. M. and L. T. Nielsen. 1951. Four new mosquito records from Utah. (Diptera, Culicidae.) Pan- Pac. Ent., 27:11-12.

Rey, J. R., Walton, W. E., Wolfe, R. J., Connelly, R., O'Connell, S. M., Berg, J., … Laderman, A. D. 2012. North American wetlands and mosquito control. *International Journal of Environmental Research and Public Health*, *9*(12), 4537–4605. http://doi.org/10.3390/ijerph9124537

Tibbets, T. M., L. Washkoviak, S.A. Tessmann, G. Jones and H.E. Copeland. 2016. Wetland Profile and Condition Assessment of the Laramie Plains Wetland Complex, Wyoming. Report to the U.S. Environmental Protection Agency. The Nature Conservancy –Wyoming Chapter, Lander, Wyoming. 46 pp. plus appendices.

Walker, E. D. and W. J. Crans. 1986. A simple method for collecting *Coquillettidia perturbans* larvae. *J Am Mosq Control Assoc*. 2(2) 239-240.

Wood, D. M., P. T. Dang, and R. A. Ellis. 1979. The mosquitoes of Canada (Diptera: Culicidae). The insects and arachnids of Canada, Part 6. Canadian Government Publishing Centre, Hull, Quebec, Canada.

TABLE 1. SPECIES OF MOSQUITOES ANTICIPATED TO OCCUR IN WYOMING (DARSIE & WARD, 2005)

Aedes cinereus	*Aedes (Ochlerotatus) punctor*
Aedes vexans	*Aedes (Ochlerotatus) schizopinax*
Aedes (Ochlerotatus) campestris	*Aedes (Ochlerotatus) sierrensis*
Aedes (Oc.) canadensis canadensis	*Aedes (Ochlerotatus) spenceri idahoensis*
Aedes (Ochlerotatus) cataphylla	*Aedes (Ochlerotatus) sticticus*
Aedes (Ochlerotatus) communis	*Aedes (Ochlerotatus) trivittatus*
Aedes (Ochlerotatus) diantaeus	*Aedes (Ochlerotatus) ventrovittis*
Aedes (Ochlerotatus) dorsalis	*Anopheles earlei*
Aedes (Ochlerotatus) euedes	*Anopheles freeborni*
Aedes (Ochlerotatus) excrucians	*Anopheles punctipennis*
Aedes (Ochlerotatus) fitchii	*Anopheles franciscanus*
Aedes (Ochlerotatus) flavescens	*Anopheles walkeri*
Aedes (Ochlerotatus) hendersoni	*Coquillettidia perturbans*
Aedes (Ochlerotatus) hexodontus	*Culex restuans*
Aedes (Ochlerotatus) impiger	*Culex pipiens*
Aedes (Ochlerotatus) implicatus	*Culex salinarius*
Aedes (Ochlerotatus) increpitus	*Culex tarsalis*
Aedes (Ochlerotatus) intrudens	*Culex territans*
Aedes (Ochlerotatus) melanimon	*Culiseta alaskaensis*
Aedes (Ochlerotatus) mercurator	*Culiseta impatiens*
Aedes (Ochlerotatus) nevadensis	*Culiseta incidens*
Aedes (Ochlerotatus) nigromaculis	*Culiseta inornata*
Aedes (Ochlerotatus) niphadopsis	*Culiseta morsitans*
Aedes (Ochlerotatus) pionips	*Psorophora signipennis*
Aedes (Ochlerotatus) pullatus	*Uranotaenia sapphirina*
Aedes (Ochlerotatus) provocans	

Wyoming Health-Related Pest Control
Category 908-K
MOSQUITO CONTROL

University of Wyoming | Extension