

5.2.1 Contact Pesticides

As the name implies, this loosely defined group of compounds is effective when mosquito larvae or pupae come in contact with it. Chemicals are absorbed through the insects outer "skin" or cuticle and may be incidentally ingested or enter the body through other routes. Contact agents can be further subdivided into two sub-groups: 1) Toxins primarily affecting an insect's nervous system and 2) toxins primarily affecting an insect's endocrine system. The nervous system agents used for mosquito larvicides in Florida during FY 94-95 include many formulations of the organophosphate compound temephos, plus one formulation of natural pyrethrum. Endocrine system agents used in this period include many s-methoprene formulations and one diflubenzuron product.

5.2.1.1 Temephos

Introduction: Organophosphate compounds (OPs) have been used for mosquito control since the early 1950s. OPs work by inhibiting the activity of cholinesterase enzymes at the neuromuscular junction, ultimately causing paralysis and death. State records indicate that the OP temephos has been used since 1969. Temephos is classified by the EPA as a General Use Pesticide and product labels carry the signal word "WARNING." Considerable research over the past decade has been conducted in Lee County by the Mote Marine Laboratory to investigate some non-target effects of using temephos. (See the section on understanding non-target effects at the back of this chapter for a discussion of the findings and a related interagency agreement for use of temephos on "Designated State Lands.")

Formulations and Dosages: Five different temephos formulations are registered with the state:

Abate 1-SG® contains 1% (wt./wt.) temephos attached to a silica (sand) granule. Rates of application vary from 5 to 20 pounds (0.05 to 0.50 lb. AI) per ac., depending upon the degree of organic content.

Abate 2-CG® contains 2% (wt./wt.) temephos attached to celatom (diatomaceous earth) grit. Rates of application vary from 2.5 to 25 pounds (0.05 to 0.50 lb. AI) per ac. depending upon the degree of polluted waters in which the product is used.

Abate 4-E® contains 44.6% (wt./wt.) active ingredient (4.0 lb./gal.), combined with a petroleum distillate and three different emulsifiers, to be diluted with water to produce a uniform spray. Rates of application vary from 0.5 to 1.5 fluid ounces per ac. (0.016 to 0.048 lb. AI/ac.).

Clarke 5% Skeeter Abate® contains 5% (wt./wt.) active ingredient imbedded in an extruded gypsum pellet. Use rates vary from 4 to 10 pounds (0.16 to 0.40 lb. AI) per ac. depending upon the degree of water pollution in which the pellet is applied. Operationally, this is considered a slow-release product with a sustained activity of three or more weeks.

Clarke Abate 5% Tire Treatment Insecticide® contains 5% (wt./wt.) temephos attached to a corncob granule. It is specifically designed to control *Aedes albopictus* (the Asian tiger mosquito), *Ae. aegypti*, and other container-breeding mosquitoes. The rate of application is 1 pound per 100 square feet of tire-pile surface area (equivalent to 21.8 lb. AI/ac.) every 30 days during the mosquito-breeding season. Tests at JAMSARL facilities in Panama City during 1995 and 1996 showed sustained control with one application for over one year.

The Abate Insecticide Technical Information Brochure PE-4090 Rev. #1 3/90 by American Cyanamid, which covers their formulations, indicates that temephos has good chemical stability in natural fresh and saline water:

The length of control achieved depends on the type of water treated and the degree of insecticide dilution after application. Applications of 1 PPM ABATE completely controlled mosquito larvae for 90 to 100 days in containers of clear water. In tidal marshes, where continual breeding and flushing occur, a single application of ABATE 4-E at 0.032 lb. AI (Active Ingredients)/acre provided adequate larval control for 10 to 14 days.

It should be noted that according to a Mote Marine Laboratory Study (Pierce et al. 1990) Abate 4E® applied in tidal marshes at 0.0032 lb. AI/ac. was still present in detectable amounts after eight hours but did not provide larval control.

Target Species: In Florida, temephos is used against virtually all mosquito species at which any control efforts are directed. In addition, some temephos formulations are occasionally used by agencies in Florida to control midges and/or sandflies as provided for under Chapter 388 F.S. and by the registered temephos labels.

Benefits: Temephos is an economical, highly effective chemical to use for a variety of mosquito species. Many formulations are available with different temephos release characteristics that enhance the overall attractiveness of this group. A nationwide 31-year track record for temephos products means that they have been and continue to be well received.

Risks/Disadvantages: Temephos is used primarily to abate floodwater mosquitoes in tidal marshes and because it is an OP, not a selective insecticide. However, temephos is one of the least toxic OPs to mammals. All temephos product labels contain the following Environmental Hazard Precautions:

This product is toxic to birds and fish. Fish and other aquatic organisms in water treated with this product may be killed. You must consult your State Fish and Game Agency before applying this product to waters or wetlands. Do not contaminate water by cleaning of equipment or disposing of wastes.

For additional information on risks specific to the Florida marine environment, see the discussion on understanding non-target effects at the back of this chapter.

5.2.1.2 Pyrethrum

Introduction: The other nervous-system contact larvicide used by mosquito control districts (MCDs) during FY 94-95 contains pyrethrum. Pyrethrum is a collective name for several natural pyrethrin compounds contained within chrysanthemums. Pyrethrins generally act on the nervous system by inhibiting the action of the sodium- and potassium-pumping mechanisms necessary for repeated transmission of impulses along nerve fibers. This action occurs rapidly upon contact, resulting first in immobility and then in death with a sufficiently high dose. Pyrethrum is classified by the EPA as a General Use Pesticide and product labels bear the signal word "CAUTION."

Pyrenone Mosquito Capsule®: This was the only pyrethrum larvicide registered for use in Florida.

It was developed in 1948 and has been manufactured by several companies over the years. When product ownership changed during 1994, the label was no longer supported. A few MCDs are now using remaining inventories.

Dosages: Pyrenone Mosquito Capsules® contain 1% (wt./wt.) pyrethrins, 10% (wt./wt.) piperonyl butoxide (PBO) acting as a synergist, plus petroleum distillates and inerts. The ingredients are contained within a red, water-soluble gelatin sphere about 3/4 inch in diameter that is wrapped with a rubber band to pressurize the contents. The design allows it to rupture, dissolve, or disintegrate when placed or thrown into water. Once the sphere wall is breached, it is propelled across the water surface by the releasing ingredients. Special oils assure a fast "spread" of the AI over water surfaces in a few minutes. Each Pyrenone Mosquito Capsule® is designed to larvicide 100 square feet at depths up to six inches, and proportionally more Pyrenone Mosquito Capsules® are recommended for greater depths.

Target Species: Pyrenone Mosquito Capsules® are labeled for use in mosquito-breeding habitats, such as stagnant pools, road and irrigation ditches, catch basins, artificial containers, lake shore lines, dairy wash lagoons, quarries, and marshy areas.

Benefits: Pyrethrum quickly dispatches both mosquito larvae and pupae. The formulation allows for easy treatment of difficult to reach spots such as containers.

Risks/Disadvantages: The product hazards statement says it:

is toxic to fish, shrimp, crabs, and other aquatic organisms. Do not apply directly to lakes, streams or ponds. May be used in mosquito-breeding areas such as marshy areas, pools, and ponds where fish, shrimp, crabs, and other desirable aquatic animals will not be harmed.

5.2.1.3 s-methoprene

S-methoprene does not produce nondiscriminatory, rapid toxic effects that are associated with nervous-system toxins. s-methoprene is a true analogue and synthetic mimic of a naturally occurring insect hormone called juvenile hormone (JH). JH is found during aquatic life stages of the mosquito and in other insects but is most prevalent during the early instars. As mosquito larva mature, the level of JH steadily declines until the 4th instar molt, when levels are very low. This is considered a sensitive period when all the physical features of the adult begin to develop. s-methoprene in the aquatic habitat can be absorbed on contact and the insect's hormone system becomes imbalanced. When this happens during the sensitive period, the imbalance interferes with 4th instar larval development.

One effect is to prevent adults from emerging. Since pupae do not eat, they eventually deplete body stores of essential nutrients and then starve to death. For these and perhaps other reasons, s-methoprene is considered an insect growth regulator (IGR).

Formulations and Dosages: Currently, five s-methoprene formulations are sold under the trade name of Altosid®. These include Altosid Liquid Larvicide (A.L.L.)® and Altosid Liquid Larvicide Concentrate®, plus solid Altosid Briquets®, Altosid XR Briquets®, and Altosid Pellets®. Early methoprene manufacturing produced equal quantities of two mirror-image molecules called r- and s-isomers. The racemic isomer (r-methoprene) is not active on mosquitoes, but is present today in

Altosid Briquets® (along with the active isomer s-methoprene) because it is part of the original EPA label package. The other products are newer or have been revised and contain only s-methoprene as their active ingredients. Altosid® labels contain the signal word "CAUTION."

Altosid Liquid Larvicide® (A.L.L.) and A.L.L. Concentrate®: These two flowable formulations have identical components except for the difference in the concentration of active ingredients. A.L.L. contains 5% (wt./wt.) s-methoprene while A.L.L. Concentrate® contains 20% (wt./wt.) s-methoprene. The balance consists of inert ingredients that encapsulate the s-methoprene, causing its slow release and retarding its ultraviolet light degradation.

Dosages: Use rates are 3.0 to 4.0 ounces of A.L.L. 5% and 0.75 to 1.0 ounce of A.L.L. Concentrate® (both equivalent to 0.01008 to 0.01344 lb. AI) per ac., mixed in water as a carrier and dispensed by spraying with conventional ground and aerial equipment. Because the specific gravity of A.L.L. is about that of water, it tends to stay near the target surface. No rate adjustment is necessary for varying water depths when treating species that breath air at the surface.

Target Species: Liquid formulations are designed to control fresh and saline floodwater mosquitoes with synchronous development patterns. Cold, cloudy weather and cool water slow the release and degradation of the active ingredient as well as the development of the mosquito larvae. Accordingly, formulation activity automatically tracks developing broods.

Altosand: This is an "On Site Granular Formulation" that end users produce by combining A.L.L. with washed sand as described in the product's labeling. Altosand Granules® were developed about 20 years ago by E. John Beidler at the Indian River MCD, primarily to control mosquitoes in densely canopied mangrove swamps and coastal salt marshes. Altosand is widely used in Florida today. Using inexpensive mixing equipment and local labor allows MCDs to save money. Altosand penetrates heavy canopies and provides outstanding control. Procuring the sand carrier locally saves freight costs. In a variation on this process, Pasco County MCD has recently combined A.L.L. and Bti granules to produce an "On Site Granular Duplex Formulation." (The benefits of duplexing the two control agents are discussed in the Stomach Toxins section below.) Both of these mixes allow A.L.L. to be applied at normal rates.

Altosid Briquets®: The Altosid Briquet® was the first solid methoprene product marketed for mosquito control beginning in 1978. It is made of plaster (calcium sulfate), 3.85 % (wt./wt.) r-methoprene, 3.85% s-methoprene (0.000458 lb. AI/briquet) and charcoal (to retard ultraviolet light degradation). Altosid Briquets® release methoprene for about 30 days under normal weather conditions.

Dosages: Application should be made at the beginning of the mosquito season and under normal weather conditions repeat treatments should be carried out at 30-day intervals. The recommended application rate is 1 Briquet per 100 square feet in non-flowing or low-flowing water up to two feet deep.

Target Species: Floodwater Aedes and Psorophora plus permanent water Anopheles, Culex, and Culiseta larvae are likely targets. Typical treatment sites include storm drains, catch basins, roadside ditches, ornamental ponds and fountains, cesspools and septic tanks, waste treatment and settlement ponds, flooded crypts, transformer vaults, abandoned swimming pools, construction, and other artificial depressions.

Altosid XR Briquets®: The XR Briquet® was approved for use in September 1988 and was first sold in Florida in 1990. It is made of hard dental plaster (calcium sulfate), 1.8% (wt./wt.) s-methoprene (0.00145 lb. AI/briquet) and charcoal (to retard ultraviolet light degradation). Despite containing only three times the AI as the "30-day briquet," the comparatively harder plaster and larger size of the XR Briquet® change the erosion rate allowing sustained s-methoprene release up to 150 days in normal weather.

Dosages: XR Briquets® should be applied 1 to 2 per 200 square feet in the lowest part of shallow depressions in no-flow or low-flow water conditions, depending on the species.

Target Species: Targets are the same as for the smaller briquet, plus *Mansonia* spp. and *Coquilletidia* spp. mosquitoes. Appropriate treatment sites for XR Briquets are the same as for the Altosid Briquets® plus cattail swamps and marshes, water-hyacinth beds, pastures, meadows, rice fields, freshwater swamps and marshes, woodland pools, flood plains, and dredge-spoil sites.

Altosid Pellets®: Altosid Pellets® were approved for use in April 1990. They contain 4% (wt./wt.) s-methoprene (0.04 lb. AI/lb.), dental plaster (calcium sulfate), and charcoal. Like the briquets discussed above, Altosid Pellets® are designed to slowly release s-methoprene as they erode. Under normal weather conditions, control can be achieved for up to 30 days.

Dosages: Label application rates range from 2.5 lbs. to 10.0 lb. per ac. (0.1 to 0.4 lb. AI/ac.), depending on the target species and/or habitat.

Target Species: The species and listed target sites are the same as listed for the briquet formulations.

Benefits: Methoprene is safe for workers and environmentally friendly. No specific safety precautions or equipment are recommended. In all situations, safety considerations and good common sense should prevail. Extended release formulations both allow end users to better use and manage the labor component of their operations and, by extending the activity period, reduce overall average daily control costs. Methoprene does not bioaccumulate. It remains effective for the prescribed period and degrades into simpler compounds. Its mode of action allows mosquito larvae to accomplish their ecological assignments and then remain present in the water for an extended time as natural food for the ecosystem's predators.

Risks/Disadvantages: An operational disadvantage of using methoprene is that the applicator cannot detect a control failure until it is too late for additional larviciding. A lack of emerging adults is the only true verification of proper treatment. In late 1986, the EPA approved the removal of the Environmental Hazards statement on solid formulation labels (Briquets®, XR Briquets®, Pellets®) which read: "Do not apply to known fish habitats".

5.2.1.4 Diflubenzuron

Introduction: Diflubenzuron is a chitin-synthesis inhibitor that acts on contact with mosquito juveniles (and potentially other chitin containing organisms) in the aquatic environment to disrupt molting processes. At the end of an insect molt, chitin is required as the hard component of the new outer skin (exoskeleton) that is necessary for biological processes, protection from the environment, and the support of internal organs. Diflubenzuron interferes with the endocrine mechanisms

(ecdysone functions) that regulate chitin production. A failure to synthesize chitin halts molting, leading to physiological difficulties, desiccation, and ultimately death. All aquatic life stages can be affected during the molting process by diflubenzuron. For these reasons, it is variously called an "insect growth inhibitor" or "insect growth regulator."

Dimilin 25W®: The only available diflubenzuron formulation for mosquito control is Dimilin 25W®, a wettable powder. The product is currently available for use in Florida as a Restricted Use Pesticide with a Special Local Need (Chapter 24c) Permit granted by the PREC in late 1991 and supported by the FDACS Bureau of Entomology and several MCDs. The toxicity of this compound to crustacea has been measured below 1 ppb and thus natural habitat applications must be avoided. Due to its toxicity to aquatic invertebrate animals, Dimilin 25W® is classified as a Restricted Use Pesticide. The Dimilin 25W® label bears the signal word "CAUTION."

Formulations and Dosages: The powder contains 25% (wt./wt.) diflubenzuron per pound. Dimilin 25W® may be diluted and applied as a wet spray or formulated on site using label instructions and broadcast as 0.25% or 0.50% sand granules. For sites other than intermittently flooded pastures, Dimilin 25W® can be applied at 3.25 ounces per ac. (0.05 lb. AI). In flooded pastures, Dimilin 25W® may be applied at 0.025 to 0.040 pound AI per ac. a maximum of six times per year, and never in floodwater that remains over 21 days.

Target Species: Dimilin 25W® can be used in freshwater sites where there is a low risk to populations of crabs, shrimp and other non-target arthropods through direct application. Such habitats include street gutters, rubber tires, storm drains, ditches and retention/detention/seepage ponds, sewage effluent and disposal fields and oxidation ponds, grassy swales, phosphate pits, tailing canals and slime ponds, wastewater biological filter beds, industrial-waste tertiary ponds and irrigation disposal fields, livestock, swine, and poultry waste lagoons, artificial ponds, channels and percolation basins designed exclusively for decorative and landscape purposes, and intermittently flooded pastures. It is not to be applied when conditions favor drift or runoff to adjacent aquatic sites.

Benefits: Dimilin 25W® is cost effective for use in "dirty water areas" where non-target organisms are not present. It has the advantage of killing the target organisms quickly (within 1 molt cycle) and thereby usually allows touch-up work in areas that may have been missed.

Risks/Disadvantages: Hay or feed for livestock should not be produced from treated pastures treated with Dimilin 25W®. Growers must be informed that the grass is not to be cut for hay. Water treated with Dimilin 25W® may not be used for irrigation or human consumption. Food or feed crops are not to be planted in treated pasture areas within six months following the last application, unless Dimilin 25W® is authorized for use on these crops. Diflubenzuron is extremely toxic to crustacea.

5.2.2 Surface Control Agents

Introduction: Larvicides in this category include oils and ethoxylated isostearyl alcohols. Unfortunately, none of the currently supported larvicides previously discussed act as pupacides. Therefore, pupal control must be achieved with these products.

Oils were first used as effective anopheline larvicides for malaria control in Florida in the early

1900s. Commonly used larviciding oils kill larvae and pupae when inhaled into the tracheae along with air at the water's surface. With low dosages (1 gal. on per ac.), they can work very slowly, taking four to seven days to give a complete kill. Higher dosage rates are usually used (up to 5 gal. per ac.) to lower the kill time. Heavy oils can suffocate but only at very high dosage rates. This oil use is not common today as it was historically. The detrimental impacts of oil treatments may actually be intensified by the elimination of many mosquito predators such as notonectids and diving beetles. The larviciding oils are probably the least studied of the mosquito larvicides.

Currently Bonide Mosquito Larvicide® and Mosquito Larvicide GB 1111® are the only two oils regularly used in Florida. Two new oil products, BVA Larvicide 2® and BVA Chrysalin Mosquito Larvae and Pupa Insecticide®, have been investigated by JAMSARL and University of California scientists. JAMSARL recently conducted BVA Larvicide 2® field trials near Tampa, Florida.

The 2 mol ethoxlate of isostearyl alcohols produces a thin (monomolecular) film on the water surface which lowers the surface tension of the water and subsequently kills mosquito larvae by inhibiting proper orientation at the "on-water" surface and/or by wetting tracheal structures and causing anoxia. Larvae normally use surface tension to suspend for long periods when breathing and/or resting. Emerging and egg-laying adults cannot be supported on the water surface when these materials are present and often drown. Currently, no alcohol-based larvicides are sold in Florida, and the last inventories of the Arosurf MSF® product are being used. This is unfortunate because the FDEP has approved Arosurf MSF® for control of pupae on "Designated State Lands." The manufacturer of the active ingredient in Arosurf MSF® is applying for a new EPA label under the trade name Agnique MMF®.

5.2.2.1 Bonide Mosquito Larvicide®:

This product is a petroleum-based "mineral oil." The "mineral oil" designation characterizes petroleum-oil-refining processes. Although it had been used in the northeastern U.S. for many years, JAMSARL scientists registered it for use in Florida in 1992 after evaluation.

Dosages: The product contains 98% (wt./wt.) mineral oil and 2% inert ingredients, including surfactants. The objective is to apply a thin film over the surface of breeding areas. For ground applications, the dosage rate is 1 to 5 gal. per ac. depending on water surface conditions and vegetative density. The label rate for aerial applications is 2 to 4 gal. per ac. also adjusted for water conditions and vegetation.

Target Species: The product is designed for surface application to intermittently flooded areas and temporary rain pools, sloughs, and log ponds. Instructions for treating salt marshes, swamps, drainage ditches, catch basins, stagnant pools, and open sewage basins are included on the label. Both larvae and pupae are targets of this product.

Benefits: Bonide Mosquito Larvicide® has good spreading characteristics and acts as both a pupacide and a larvicide.

Risks/Disadvantages: This oil, like the others discussed in this chapter, produces a thin "oil slick" on the water surface when applied. When viewed under some lighting conditions, the resulting unnatural appearance may be objectionable, precluding widespread use of the oil in some areas.

5.2.2.2 Mosquito Larvicide GB-1111®

This product is a petroleum based "naphthenic oil." The "naphthenic oil" designation characterizes petroleum-oil-refining processes. The product is most often referred to as Golden Bear 1111® or simply GB-1111. Another mosquito control product, GB-1356, was nearly identical to GB1111, but label support was withdrawn in the early 1990s. Agencies are using remaining inventories of GB-1356. Product use of the two has been combined for this report. Both labels contain the signal word "CAUTION."

Dosages: GB-1111 contains 99% (wt./wt.) oil and 1% (wt./wt.) inert ingredients including an emulsifier. The nominal dosage rate is 3 gal. per ac. or less. Under special circumstances, such as when treating areas with high organic content, up to 5 gal. per ac. may be used.

Target Species: GB-1111 is effective on a wide range of mosquito species. Applied to breeding areas, GB-1111 is an effective material against any mosquito larvae and pupae obtaining atmospheric oxygen at the water surface. It can even be effective in treating adult mosquitoes as they emerge.

Benefits: GB-1111 is a material with good spreading characteristics. A big benefit of having it at a MCD's disposal is the fact that it acts as both a pupacide and a larvicide. Even emerging adults can be affected.

Risks/Disadvantages: This oil, like the others discussed in this chapter, produces a thin "oil slick" on the water surface when applied. When viewed under some lighting conditions, the resulting unnatural appearance may be objectionable, precluding widespread use of the oil in some areas.

5.2.2.3 BVA Larvicide 2®

This product is new and EPA registration has been applied for. It was recently field and laboratory tested in Florida. BVA Larvicide 2® contains 97% distilled petroleum oil and 3% inert ingredients. It is formulated with a structurally modified mineral oil to produce a water-clear product designed to work fast and effectively as both a larvicide and a pupacide in mosquito-breeding areas.

Formulations and Dosage: Application rates call for 3 to 5 gal. per ac. Where vegetation is not a factor, 3 gal. per ac. should be sufficient. If vegetation is dense, the higher application rate will be necessary.

Target Species: BVA Larvicide 2® is designed to be effective against a variety of mosquito species. Targets include larvae, pupae, and emerging adults.

Benefits: BVA Larvicide 2® has good spreading characteristics. Its water-clear character is considered advantageous when dealing with environmentally concerned citizens.

Risks/Disadvantages: This oil, like the others discussed in this chapter, produces a thin "oil slick" on the water surface when applied. When viewed under some lighting conditions, the resulting unnatural appearance may be objectionable, precluding widespread use of the oil in some areas.

5.2.2.4 BVA Chrysalin Mosquito Larvae and Pupae Insecticide®

This also is a brand-new product about which little is known. It is a lightweight (6.82 lb./gal.) larvicide containing mineral oil (22.94% wt./wt.), a botanically derived larvicide (0.024 % wt./wt. Dihydro-5-pentyl-2(3H)-furanone and 0.049% wt./wt. Dihydro-5-heptyl-2(3H)-furanone), and 72.97% inert petroleum distillates. In addition, it contains 4.015% d'Limonene, which gives it a pleasant citrus odor. It is intended for use by MCDs, homeowners, and farmers. The label precautionary statement bears the signal word "CAUTION."

Formulations and Dosages: BVA Chrysalin® can be applied without further dilution or use of any sophisticated spray equipment. When mixed with water, it forms a quick-breaking spray emulsion. Application rates call for 2 to 5 gal. per ac. as a course spray. In areas of dense vegetation, the higher rate should be used. When applied as directed, highest mortality is achieved within 24 hours of application under normal conditions. Label recommendations include re-application at minimum rates after 14 days in waters in which fish and wildlife abound.

Target Species: BVA Chrysalin® is designed to be effective against larvae, pupae, and emerging adults of a variety of mosquito species. Mosquito habitats include but are not limited to swamps, ditches, tires, pools, fountains, ponds and areas around homes and farmyards.

Benefits: The product may work up to three weeks. Tests on two Culex species in California experimental ponds showed effects at up to 21 days after one treatment. Its widespread use by home and farmers could lead to reductions in domestic service requests for MCDs. The citrus odor is pleasant to work around and may help dispel potential fears of environmentally conscious citizens.

Risks/Disadvantages: Applicators must wait 14 days before re-applications and then use only minimum doses. Users must avoid applications to artificial surfaces sensitive to mineral oils.

5.2.2.5 Arosurf MSF®

Arosurf MSF® is 100% (wt./wt.) the two mol ethoxylate of isostearyl alcohol [poly(oxy-1,2-ethanediyl), alpha-isooctadecyl-omega-hydroxyl]. It belongs to a group of surfactants that have been routinely used in detergents and cosmetics for over 20 years. It is a biodegradable, non-ionic, surface control material that spontaneously and rapidly spreads over the surface of the water to form an ultra-thin film that is about one molecule in thickness.

Monomolecular surface films do not kill by toxic action but exert a physical impact on mosquito populations that cause larvae, pupae, and emerging adults to drown. They act by slightly reducing the strength of the water's surface. They affect only species that depend on the air-water interface, such as mosquitoes. The weakened tension of the water's surface fails to support larvae and pupae and allows water to wet their breathing apparatus so they drown. This product is a registered larvicide and pupicide with a currently active EPA label. It is not currently formulated.

Dosages: No mixing is required. It may be applied directly from its container using hand or power sprayers. Application rates for this mosquito larvicide and pupicide generally range from 0.2 to 0.5 gal. per surface ac. of water.

Target Species: It is effective against most species. It may be used safely in potable waters, waters bearing fish and other aquatic organisms, and in runoff waters that enter fish-bearing waters. Mosquitoes that require little or no surface contact for breathing (*Mansonia* spp., *Coquellitidia*

alone or in combination to destroy the gut wall. This leads to paralysis and death of the larvae.

spp., *Culex pilosis*, *Cx. erraticus*) require properly timed applications at sensitive, surface-contacting stages (pupae to emerging adult) for maximum impact.

Benefits: Because the method of action is physical rather than toxic or biochemical, mosquitoes cannot easily develop resistance. It is especially useful against chemically resistant species. It decomposes into harmless elements. It has a broad range of activity. It kills all stages and can entrap and drown egg-laying females or surface-resting males.

Risks/Disadvantages: Arosurf MSF® is nearly invisible on the surface of the water. Testing for its presence requires placing a few drops of ADOL indicator oil on the surface and checking for a reaction, a time-consuming process. Dried vegetation and floating debris tends to absorb the product, and sustained winds tend to make debris pile up in localized areas.

5.2.2.6 Agnique MMF 5996®

Agnique MMF 5996® is an experimental monomolecular surface film that is chemically identical to Arosurf MSF®. Testing is being conducted by Lee County MCD scientists who developed Arosurf MSF®. The safety, properties, application rates, and mosquito-controlling efficacy of Agnique MMF 5996® have been shown to be comparable and in some cases superior to that of Arosurf MSF®.

5.2.3 Stomach Toxins

Introduction: Mosquito control has two stomach toxins whose active ingredients are manufactured by bacteria. These control agents are often designated as bacterial larvicides. Their mode of action requires that they be ingested to be effective, which can make them more difficult to use than the contact toxins and surface control agents.

Bacteria are single-celled parasitic or saprophytic microorganisms that exhibit both plant and animal properties and range from harmless and beneficial to intensely virulent and lethal. A beneficial form, *Bacillus thuringiensis* (Bt), is the most widely used agricultural microbial pesticide in the world. It was originally isolated from natural Lepidopteran (butterflies and moths) die-offs in Germany and Japan. Various Bt products have been available since the 1950s, and in 1976, Dr. Joel Margalit and Mr. Leonard Goldberg isolated from a stagnant riverbed pool in Israel a subspecies of *B. thuringiensis* that had excellent mosquito larvicide activities. It was named Bt variety israelensis (Bti) and later designated *Bacillus thuringiensis* Serotype H-14. Either of these two designations may be found on the labels of many bacterial mosquito larvicide formulations used today. Another species of bacteria, *B. sphaericus*, also exhibits mosquito larvicide properties.

5.2.3.1 *Bacillus thuringiensis israelensis* (Bti)

Introduction: Like a tiny chemical factory capable of only one production run, each Bti organism may produce, if the environmental conditions around it are favorable, five different microscopic protein pro-toxins packaged inside one larger protein container or crystal. The crystal is commonly referred to as delta (δ)-endotoxin. If the δ-endotoxin is ingested, these five proteins are released in the alkaline environment of an insect larva's gut. The five proteins are then converted into five different toxins if specific enzymes also are present in the gut. Once converted, these toxins work alone or in combination to destroy the gut wall. This leads to paralysis and death of the larvae.

Bti is grown commercially in large fermentation vats using sophisticated techniques to control environmental variables such as temperature, moisture, oxygen, pH, and nutrients. The process is similar to the production of beer, except that Bti bacteria are grown on high-protein substrates such as fish meal or soy flour, and the spore and delta endotoxin are the end products. At the end of the fermentation process, Bti bacteria exhaust the nutrients in the fermentation machine, producing spores before they lyse and break apart. Coincidental with sporulation, the delta endotoxin is produced. The spores and delta endotoxins are then concentrated via centrifugation and microfiltration of the slurry. It can then either be dried for processing and packaging as a solid formulation(s) or further processed as a liquid formulation(s). Since some fermentation medium (e.g. fishmeal) is always present in liquid formulations, they generally smell somewhat like the medium.

Formulations and Dosages: There are five basic Bti formulations available for use: Liquids, powders, granules, pellets, and briquets. Liquids, produced directly from a concentrated fermentation slurry, tend to have uniformly small (2-10 micron) particle sizes which are suitable for ingestion by mosquito larvae. Powders, in contrast to liquids, may not always have a uniformly small particle size. Clumping, resulting in larger sizes and heavier weights, can cause particles to settle out of the feeding zone of some target mosquito larvae, preventing their ingestion as a food item. Powders must be tank-mixed before application to an inert carrier or to the larval habitat, and it may be necessary to mix them thoroughly to achieve a uniformly small consistency. Bti granules, pellets, and briquets are formulated from Bti primary powders and an inert carrier. Bti labels contain the signal word "CAUTION."

Since 4th instar mosquito larvae quit feeding prior to becoming pupae, it is necessary to apply Bti prior to this point in their development. Although the details are poorly understood, evidence suggests that larvae also undergo a period of reduced feeding or inactivity prior to molting from 1st to 2nd, 2nd to 3rd, and 3rd to 4th instars. If Bti is applied at these points in their development, the toxic crystals may settle out before the larvae resume feeding, and with synchronous broods of mosquitoes, complete control failures may result. With asynchronous broods, efficacy may be reduced. Kills are usually observed within 24 hours of toxin ingestion. As a practical matter, apparent failures are usually followed up with oil treatments.

The amount of toxins contained within Bti products are reported indirectly as the result of at least two different bioassays and are difficult to equate to one another. Prepared volumes of toxins are applied to living mosquito larvae and the resulting mortality produces through formulae numerical measures known as International Toxic Units (ITUs) and *Aedes aegypti* International Toxic Units (AA-ITUs). These measures are only roughly related to observed efficacy in the field and are therefore inappropriate to consolidate and report on like other toxicants.

Bti Liquids: Currently, three commercial brands of Bti liquids are available: Aquabac XT®, Teknar HP-D®, and Vectobac 12AS®.

Dosages and Formulations: Labels for all three products recommend using 4 to 16 liquid oz./ac. in unpolluted, low-organic water with low populations of early instar larvae (collectively referred to below as clean water situations). The Aquabac XT® and Vectobac 12AS® (but not Teknar HP-D®) labels also recommend increasing the range from 16 to 32 liquid oz./ac. when late 3rd or early 4th instar larvae predominate, larval populations are high, water is heavily polluted, and/or algae are abundant. The recommendation to increase dosages in these instances (collectively referred to below as dirty water situations) also is seen in various combinations on the labels for all

other Bti formulations discussed below.

Duplex: Bti liquid may also be "Duplexed" with the Altosid Liquid Larvicide® discussed above. Because Bti is a stomach toxin and lethal dosages are somewhat proportional to a mosquito larvae's body size, earlier instars need to eat fewer toxic crystals to be adversely affected. Combining Bti with methoprene (which is most effective when larvae are the oldest and largest) allows MCDs to use less of each product than if using only one product. However, since A.L.L. by itself has such a long residual activity, "duplexing" is not widely practiced. Financially, most savings are realized for treatments of mosquitoes with long larval development periods and asynchronous broods.

Bti Powders: Aquabac Primary Powder®, Vectobac TP® and Bactimos WP® brands of Bti powders are available. The Vectobac TP® label recommends using a calculated 3.2 to 6.4 oz. (by weight)/ac. in clean water, and up to 12.8 oz./ac. in dirty water situations. The Bactimos WP® label correspondingly recommends using 2 to 6 oz./ac. and up to 12 oz./ac.. Aquabac Primary Powder® currently is labeled for manufacturing use only. However, the label is currently being amended by the EPA to allow end-user applications in quantities similar to those of the other powder formulations.

Bti Sand Granules: Until the latter part of 1996, commercial formulations of Bti sand granules were not available. However, labeling was available for both Vectobac® and Bactimos Bti® powders to guide end users in making their own "On-Site Sand Granules." Sand formulations require coating the particles with an oil, such as GB-1111 or Bonide Mosquito Larvicide®, and then applying dry Bti powder which will stick to the oil. In Florida, most target mosquito species graze the water surface or the interior of the water column, and not the bottom. It is desirable to stick the powder to the sand in a way that Bti is released upon contact with the water and thus "puts the food on the table" for the larvae.

Bti Corncob Granules: Granular formulations use a carrier that is dense enough to penetrate heavy vegetation. There are currently two popular corncob granule sizes used in commercial formulations. Aquabac 200G®, Bactimos G®, and Vectobac G® are made with 5/8 grit-crushed cob, while Aquabac 200 CG® (Custom Granules) and Vectobac CG® are made with 10/14 grit cob. Aquabac 200 CG® is available by special request. The 5/8 grit is much larger and contains fewer granules per pound. The current labels of all Bti granules recommend using 2.5 to 10 lb./ac. in clean water and 10 to 20 lb./ac. in dirty water situations.

Bti Pellets: Bactimos Pellets® are the only extruded Bti product on the market today. They are manufactured using a larval food as the Bti carrier, and the manufacturer claims that this helps attract feeding larvae. Bactimos Pellets® contain twice the amount of toxic units as Bactimos Granules® (corncob), and the label correspondingly recommends using only half as much by weight in both clean water and dirty water situations.

Bti Briquets (donuts): Bti donuts are a sole-source product. They are a mixture of Bti, additives, and cork. They are designed to float and slowly release Bti particles for up to 30 days. They apparently are attractive to raccoons and possibly other wildlife because of their odor, and may sometimes be disturbed or carried off. Donuts may be staked in place to prevent wind from moving them from a site's littoral zone into open water. The use rate is one donut per 100 square feet in clean water and up to four donuts per 100 square feet in dirty water.

Target Species: Bti adversely affects larval stages of insect species in the Order Diptera, Suborder

Nematocera, Families Culicidae (mosquitoes) and Simuliidae (black flies). Bti has been shown effective for numerous mosquito species, including members of the mosquito genera *Aedes*, *Anopheles*, *Culex*, *Culiseta*, and *Psorophora* commonly targeted in Florida.

Benefits: Products containing Bti are ideally suited for Integrated Pest Management programs because the active ingredient does not interrupt activities of most beneficial insects and predators. Since Bti has a highly specific mode of action, it is an insecticide of minimal environmental concern. Bti controls all larval instars provided the insects have not quit feeding, and can be used in almost any aquatic habitat with no restrictions. It may be applied to irrigation water and any other water sites except treated finished drinking water. Bti is fast acting and its efficacy can be evaluated almost immediately. It usually kills larvae within one hour after ingestion, and since each instar must eat in order for the larvae to grow, that means Bti usually kills mosquito larvae within 24 hours of application in Florida. It leaves no residues, and it is quickly biodegraded. Resistance is unlikely to develop simultaneously to the five different toxins derived from the Bti delta-endotoxin since they have five different modes of action. This suggests that this mosquito larvicide will continue to be effective for many years.

Risks/Disadvantages: Bti labels carry the CAUTION signal word, suggesting the material may be harmful if inhaled or absorbed through the skin. However, the four-hour Inhalation LC 50 in rats is calculated to be greater than 2.1 mg/liter (actual) of air, the maximum attainable concentration. The acute Dermal LD 50 in rabbits is greater than 2,000 mg/kg body weight and is considered to be nonirritating to the eye or skin. That is equivalent to a 220 pound individual spilling more than a half gallon of Bti liquid onto the body or into the eyes. Toxicology profiles also suggest that the inert ingredients (not the Bti.) in liquid formulations may cause minor eye irritations in humans. The acute Oral LD 50 in rats is greater than 5,000 mg/kg body weight (similar to an individual drinking over five quarts), suggesting the material is practically nontoxic in single doses. Common table salt has an LD 50 of 4,000 mg/kg of body weight.

Bti applied at label rates has virtually no adverse effects on applicators, livestock, or wildlife, including beneficial insects, annelid worms, flatworms, crustaceans, mollusks, fish, amphibians, reptiles, birds, and mammals. However, non-target activity on larvae of insect species normally associated with mosquito larvae in aquatic habitats has been observed in larvae in the Order Diptera, Suborder Nematocera, Families Chironomidae (Midges), Ceratopogonidae (Biting Midges), and Dixidae (Dixid Midges). These non-target insect species, taxonomically closely related to mosquitoes and black flies, apparently contain the necessary gut pH and enzymes to activate delta-endotoxins. However, the concentration of Bti required to cause these effects is 10 to 1,000 times higher than normal use rates.

Concerning the use of Bti, timing of application is extremely important. Optimal benefits are obtained when treating 2nd or 3rd instar larvae. Treatments at other development stages may provide less than desired results. Therefore, a disadvantage of using Bti is the limited treatment window available.

5.2.3.2 *Bacillus sphaericus* (Bs)

Introduction: *Bacillus sphaericus* is a commonly occurring spore-forming bacterium found throughout the world in soil and aquatic environments. Some strains produce a protein endotoxin at the time of sporulation. It is grown in fermentation vats and formulated for end use with processes similar to that of Bti. A standard bioassay similar to that used for Bti has been developed to

determine preparation potencies. The bioassay uses *Cx. quinquefasciatus* 3rd to 4th instar larvae. The endotoxin destroys the insect's gut similarly to Bti and has been shown to have activity against larvae of many mosquito genera such as *Aedes*, *Culex*, *Culiseta*, and *Psorophora*. The toxin is active only against the feeding larval stages and must be partially digested before it becomes activated. At present, the molecular action of Bs is unknown. Isolation and identification of the primary toxin responsible for larval activity has demonstrated that it is a protein with a molecular weight of 43 to 55 kD.

VectoLex CG®: VectoLex-CG® is the trade name for a granular formulation of Bs (strain 2362). The product has a potency of 50 BSITU/mg (Bs International Units/mg) and is formulated on a 10/14-mesh ground corncob carrier. The VectoLex-CG® label carries the "WARNING" hazard classification.

Dosages: VectoLex-CG® is designed to be applied by ground (by hand or truck-mounted blower) or aerially at rates of 5-10 lb./ac.. Best results are obtained when applications are made to larvae in the 1st to 3rd instars. Use of the highest rate is recommended for dense larval populations. Larval mortality may be observed as soon as a few hours after ingestion but typically takes as long as 2-3 days, depending upon dosage and ambient temperature. VectoLex-G® should be stored in a cool, dry place in an intact product package. Once the VectoLex-G® package is opened, moisture can be absorbed by the product, leading to loss of activity over time. Refrigeration is not necessary.

Target Species: *Bacillus sphaericus* adversely affects larval stages of insect species in the Order Diptera, Suborder Nematocera, Family Culicidae (Mosquitoes). *Culex* species are the most sensitive to Bs, followed by *Anopheles* and some *Aedes* species. In Florida, *Culex* spp. and *Anopheles* spp. may be effectively controlled. Several species of *Aedes*, including *Ae. aegypti* and *Ae. albopictus*, have shown little or no susceptibility, and salt-marsh *Aedes* species are not susceptible. *Bacillus sphaericus*, in contrast to Bti, is virtually nontoxic to black flies (Simuliidae).

Benefits: *Bacillus sphaericus* has demonstrated the unique property of being able to control mosquito larvae in highly organic aquatic environments, including sewage-waste lagoons, animal-waste ponds, and septic ditches. After a single application at labeled rates, extensive field evaluations have shown VectoLex-CG® to persist for 2-4 weeks. Field evaluations with VectoLex-CG® have shown that Bs may undergo limited recycling in certain organically rich environments.

Risks/Disadvantages: VectoLex-CG® has been extensively tested and has had no adverse effects on mammals or non-target organisms. *Bacillus sphaericus* technical material was not infective or pathogenic when administered as a single oral, intravenous, or intratracheal installation in rats. No mortalities or treatment-related evidence of toxicologic effects were observed. The acute oral and dermal LD 50 values are greater than 5000 mg/kg and greater than 2000 mg/kg, respectively. The technical material is moderately irritating to the skin and eye. Oral exposure of Bs is practically nontoxic to mallard ducks. No mortalities or signs of toxicity occurred following a 9000 mg/kg oral treatment. Birds fed diets containing 20% wt./wt. of the technical material experienced no apparent pathogenic or toxic effects during a 30-day treatment period. Mallards given an intraperitoneal injection of Bs demonstrated toxicologic effects including hypoactivity, tremors, ataxia, and emaciation. The LD 50 value was greater than 1.5 mg/kg. Although viable spores were cultured from treated birds, replication was not evident, since low numbers of cultured organisms were reported.

The parasitic nematode *Romanomermis*, the pathogenic protozoan *Nosema algerae*, and some non-digestible algae have been examined as biocontrol agents by university, government (USDA), and by mosquito control organizations. Thus far, results have been mixed. In some cases, the application techniques that have been used need additional development.

5.2.4.3 Copepods

Another group of biocontrol agents with promise for mosquito control is the predacious copepods. In Florida, season-long control was achieved in trials in tire habitats in Florida. Copepods are easy to rear and to deliver to target sites in the field, and they perform well when used with insecticides.

5.2.4.4 Predacious Mosquitoes

The predacious mosquito larvae in the genus *Toxorhynchites* have been studied in a variety of urban areas as control agents for other container inhabitants such as *Ae. albopictus*. The idea is to release captive-bred adults who will lay their eggs in target habitats. Their growing larvae will then eat the larvae of target species. *Toxorhynchites* spp. appear to affect other mosquito species that develop in tree holes. However, their introduction into urban environments to control nuisance and disease vectoring mosquitoes that breed in container habitats has proven unsuccessful.

5.2.4.5 Predacious Fish

Fish are the most extensively used larval biocontrol agent. Florida MCDs do not stock fish extensively as is done in California. *Gambusia* spp. are the most desirable larvivorous fish. Fish are usually just collected by seine, net, or trap from one location and transported to another for stocking. Since most field personnel are not trained taxonomists, these fish collections are likely not to be sorted before transport to a new site and usually contain various species. Escambia County mosquito control workers seed abandoned swimming pools in Pensacola with *Gambusia* spp. and then use the sites as their stock sources. A handful of control programs either purchase their *Gambusia* from fish farms or actually raise them on their property with aerated tanks, usually made from cracked but repairable concrete crypts which are readily available as donations from local funeral homes. The fish are placed in a variety of permanent and semi-permanent water habitats as needed. These habitats are usually those that are seasonally dry and are often man made. *Rivulus* spp. fish currently are being evaluated in saltwater habitats, especially in Brevard County.

Benefits: One advantage of some biocontrol agents is host-specificity. This factor affords minimal disturbance to non-target species and to the environment.

Risks/Disadvantages: Ironically, this specificity also deters commercialization and application of some biocontrol agents. Industry is faced with a generally narrow market, increased capital outlays, and the need to train their sales force and end users.

5.3 RECENT LARVICIDE USE

Forty (80%) of 50 MCDs who reported to FDACS during FY 94-95 used larvicides. Fifteen of the them larvicided by air, and all 40 larvicided by ground. The 15 MCDs treated a combined total of 358,049 acres by air, at an average rate of 23,870 acres, with a range of 1096 to 173,915 acres. They also treated an average of 3478 acres (range 3 to 15,111) on the ground. Five of them

Acute aquatic freshwater-fish toxicity tests were done on bluegill sunfish, rainbow trout and daphnids. The 96-hour LC 50 and NOEC value for bluegill sunfish and rainbow trout was greater than 15.5 mg/L.; the 48 hour EC 50 and NOEC value for daphnids was greater than 15.5 mg/L. Acute aquatic saltwater-fish toxicity tests were done on sheep head minnows, shrimp, and oysters. The 96- hour LC 50 value for both sheep head minnows and shrimp was 71 mg/L., while the NOEC value was 22 mg/L. for sheep head minnows and 50 mg/L. for shrimp. The 96-hour EC 50 value for oysters was 42 mg/L. with a NOEC of 15 mg/L.

Invertebrate toxicity tests were done on mayfly larvae and honeybees. The LC 50 and NOEC value for mayfly larvae was 15.5 mg/L. Honeybees exposed to 10E4-10E8 spores/ml for up to 28 days demonstrated no significant decrease in survival when compared to controls. Acute toxicity of *B. sphaericus* to non-target plants was evaluated in green algae. The 120-hour EC 50 and NOEC values were greater than 212 mg/L.

Bacillus sphaericus will not regenerate in salt water, rendering its use impractical for control of saltwater mosquitoes. Cycling is limited to permanent freshwater bodies, and if organics are very high, recycling may be minimal.

5.2.4 Natural Agents

Introduction: This group of larvicides is comprised of biological organisms. The use of biological organisms to combat pest insects, such as mosquitoes, is termed biological control or biocontrol. Generally, this definition includes natural and genetically modified organisms and means that the agent must be alive and able to attack the mosquito. The overall premise is simple: biocontrol agents that affect mosquitoes naturally are reared artificially and then released into the environment, usually in far greater numbers than they normally occur, and often in habitats that previously were devoid of them. Research on predators of mosquitoes, such as fish, dragonflies, and frogs is ongoing. Currently, there is no small- or large-scale program for application developments of these organisms. As the development of mosquito control technology moves forward, new biocontrol agents will be discovered. Ultimately, it is hoped that biological control will become an integral part of Florida's mosquito control programs.

Biocontrol is not a magic bullet for mosquito control in the 21st century. It should be considered a set of tools that a mosquito control program can use when it is economically feasible. When combined with conventional chemicals and cultural control procedures, biocontrol agents can provide short, and, occasionally, long-term control. Biocontrol as a conventional control method should aim at the weakest link of the life cycle of the mosquito. In most cases, this is the larval life stage.

At least ten natural control agents have been tried or are currently used by major mosquito control programs in Florida. These include non-digestible algae, a water mold (*Lagenidium giganteum*), a bacterium (Bs), a pathogenic protozoan (*Nosema algerae*), parasitic nematodes (*Romanomermis* spp.), predacious copepods, predatory mosquitoes (*Toxorhynchites* spp.), and fish (*Gambusia* spp. plus others) which may be used as larvicides. Bats and purple martins (*Progne subis*) are biocontrol agents that may be used as adulticides, a pesticide group discussed later in this report. Recently, Bs (see above under "Stomach Toxins") and *L. giganteum* were registered with the EPA as mosquito larvicides. With the exception of fish, which are used at least occasionally by nearly all the 50 reporting programs, none of these biocontrol agents is used operationally. All of the

biocontrol agents mentioned have been field or lab tested in Florida.

5.2.4.1 *Lagenidium giganteum*

Lagenidium giganteum is a parasitic, yeast-like water mold or fungus. The life cycle begins with a motile zoospore (asexual stage) that seeks out mosquito larvae, attaching to and penetrating the cuticle via a germ tube. The fungus grows inward, eventually filling the body cavity and killing the mosquito larva. The fungus can then be released from the infected cadaver, generating more zoospores that can infect other larvae. The sexual cycle produces oospores that can maintain the fungus during unfavorable conditions, such as long periods of drought. Upon flooding, oospores release infective zoospores to start the cycle again.

Liginex AS®: It is EPA registered (No. 69592-Y) as an aqueous suspension with the trade name Liginex AS®. The product label contains the signal word "CAUTION."

Dosages: Liginex AS® contains 40% (wt./wt.) *L. giganteum* (California strain) mycelium (1010 CFU or Colony Forming Units, a concentration measure by cell counts per L.) and 60% inert ingredients. Liginex AS® may be applied from ground or air. Label rates range from 9 to 180 fluid ounces (fl. oz.) per ac.. Most treatments will require 20 to 80 fl. oz./ac., a common rate being 25 fl. oz./ac.. Zoospores form within 16 hours following application and mortality occurs within 24 to 48 hours.

Target Species: It is intended for use in unpolluted water with low organic loads and low salinity levels. It is labeled for use in drainage ditches, flood plains, wildlife ponds, small containers, margins of rivers and streams, and marshes. Crop uses include rice, soybeans, and irrigated pastures. Susceptible genera include *Aedes*, *Anopheles*, *Coquilletidia*, *Culex*, *Culiseta*, *Deinocerites*, *Eretmapodites*, *Haemagogus*, *Mansonia*, *Opifex*, *Orthopodomyia*, *Psorophora*, *Sabethes*, *Uranotaenia*, and *Wyeomyia*.

Benefits: Because Liginex AS® is very host specific, studies have demonstrated no mammalian or non-mammalian toxicity. Because the fungus recycles in the mosquito, effective control can last for three to four weeks or more. Total applications in a season can be reduced in permanent or semi-permanent bodies of water with continuously breeding mosquitoes.

Risks/Disadvantages: Because Liginex AS® is a living product, it has a shorter shelf life than chemical pesticides. Because infection does not result in immediate mortality, it requires an understanding of how fungus works to accurately evaluate population reduction. Liginex AS® is not effective in highly polluted water, water with high levels of organic matter, or water with high salinity. The label environmental hazards statement indicates:

Do not discharge effluent containing this product into lakes, streams, ponds, estuaries, oceans or public waters unless this product is specifically identified and addressed in an NPDES permit. Do not discharge effluent containing this product to sewer systems without previously notifying the sewage treatment plant authority. For guidance, contact your State Water Board or Regional Office of the EPA.

5.2.4.2 Nematodes, Protozoans, and Algae

larvicided another 9173 acres, but their reports list combined figures for air and ground treatments. Together, the 15 MCDs treated 418,387 (91.2%) of the 458,937 total acres larvicided. The other 25 MCDs larvicided the balance of 39,549 acres at an average rate of 1582 acres, range 3 to 13,495 acres each. Except for Leon County, MCDs that larvicide by air are in coastal peninsular Florida. In contrast, ground-larviciding MCDs are spread throughout the state.

As shown in Appendix V, 19 different larvicides were used during FY 94-95. The average treatment required about 1.0 lb. AI/ac. Oil use biases this figure upward. Oils are nearly all AI and are used at an average rate of over 20 lb./ac. In contrast, the average endocrine-based contact toxin, use was about 0.016 lb. AI/ac.. What is important is the concentration of AI used. All MCDs were within or below ranges stated on EPA product labels. Larvicide inert ingredients are not included in the figures, but they also were applied to target sites. For example, Abate 4E® contains 40% wt./wt. (about 3.59 lb./gal.) Exxon oil (CAS. No. 008002-05-9), and about 5909 pounds were applied at 0.027 lb./ac. along with the AI. Likewise, Altosid Briquet®, XR Briquet®, and Pellet® users applied about 12,850 pounds of charcoal and calcium sulfate at an average rate of 2.28 lb./ac. Altosid Briquet® users also applied about 202 pounds (0.06 lb./ac.) of methoprene.

Contact toxins were used for 66.7% (111,047 acres) of the treatments. Temephos was used to treat over two and one half times as many acres as those treated with s-methoprene (not counting "Duplex" mixes). Diflubenzuron and Pyrethrum were seldom used and accounted for a total of only 151.4 acres (0.03%). Stomach toxins (Bti formulations) administered without s-methoprene were the next most popular group and accounted for 24.0 % (306,024 acres) of the larviciding activity. Surface active agents were used for 5.0% (22,753 acres) of the applications, while "Duplex" mixes (contact toxin-stomach toxin combinations of s-methoprene and Bti) were used for 4.2% (19,113 acres) of the treatments. Biocontrol agent larviciding with predacious fish applications or with predacious Toxorhynchites spp. mosquito releases were not itemized by MCDs for FDACS during the fiscal year.

When measured by the number of end users, delta-endotoxin was the most popular larvicide, with 34 (85%) of the 40 MCDs reporting the use of Bti formulations. Thirty (75%) used s-methoprene, 26 (65%) used oils, 18 (45%) used temephos, while Arosurf MSF®, Pyrenone Capsules®, Dimilin 25W®, and "Duplex" were each applied by 15% or fewer of the larviciding MCDs. This is not to say that the less frequently used formulations are not important. Most MCDs use a variety of larvicides, but have one or two favorites. During FY 94-95, the 40 MCDs used an average of five larvicides each (range 1 to 9). (See Appendix VI.)

5.3.1 Equipment Available