

AQUABAC Mosquito Biolarvicide Technical Bulletin

AQUABAC is a microbial larvicide containing *Bacillus thuringiensis* subspecies *israelensis* (Bti or Bt H-14), a naturally occurring soil bacterium that produces protein toxins (insecticidal crystal protein - ICP) and a spore. The insecticidal activity of AQUABAC formulations and all Bti formulations is due to the presence of the protein toxins. The spore has no effect on the larvicide activity.

AQUABAC formulations are unique and specifically manufactured to enhance larvicidal effects of the protein toxin and to ensure the toxin's biological stability.

Two formulations of AQUABAC are available from constantly replenished stock:

AQUABAC xt Biological Larvicide Aqueous Suspension 1,200 ITU/Mg,

AQUABAC (200G) Corn Cob Granule 200 ITU/Mg (5/8 and 10/14 mesh available),

The potency of AQUABAC is controlled within specific limits using a bioassay procedure based on toxicity to larvae of *Aedes aegypti*. Quality control bioassays are conducted prior to release of each lot product. Other laboratories conduct analyses on each lot to assure there are no microbial contaminants present in the formulations.

AQUABAC xt Biological Larvicide Aqueous suspension is available in:

- **2.5 gallon (9.465-lt) plastic bottles, packaged 2 bottles per carton**
- **30 gallon (113.56-lt) HDPE Drums**

AQUABAC 200G is available in

- **40 pound (18.14-kg) poly bags, 40 bags per standard shipping pallet**
- **1,200 pound bulk sacks**

MODE OF ACTION

Mosquito larvae are killed by ingesting the protein crystal (delta endotoxin). The crystal is broken down in the larval midgut by a combination of enzymes and the alkaline gut environment. The epithelium of the midgut is destroyed resulting in gut paralysis, complete loss of ionic regulation between the midgut and hemocoel, and larval death. Because the AQUABAC must be ingested to be effective, the products do not affect pupae, adults, or late larval stages when active feeding has ceased.

Death of larvae is rapid, often occurring within one hour at field use rates. Complete mortality occurs in the laboratory within 24 hours. AQUABAC xt is specifically formulated to enhance its effects on mosquito larvae. Particle size is small, averaging 2 to 10 microns, but a significant amount of the formulated crystal protein is in agglomerated form so that ULV deposition is enhanced. These particle size characteristics improve both ground and aerial applications.

EFFECT ON NON-TARGET ORGANISMS

ENVIRONMENTAL FATE:

AQUABAC Bti is naturally occurring and safe to the environment. It shows no cross resistance to chemical larvicides and is especially well suited for use in areas where there is demonstrated resistance to organophosphates or pyrethroids.

AQUABAC does not persist in soil or water. The protein crystal separates from the aquatic environment and settles to the bottom. They are deactivated by becoming food for other microorganisms or being absorbed into soil or other organic particles. Spores do not recycle or regenerate in the field.

INVERTEBRATE TOXICOLOGY:

Bacillus thuringiensis subspecies *israelensis* (Bti) has no toxic effects on beneficial insects such as lady bird beetles, honeybees, mayflies, dragonflies, damselflies, stoneflies, caddisflies, and true bugs. Among Diptera *Chaoborous* species *Ephydra riparia*, *Musca domestica*, *Odontomyia* species, and *Polpedilum* species are not susceptible to Bti. Some mortality occurs among *Chironomous pulmosus*, *Chironomous stigmaterus*, *Dixa* species, *Goeldichironomous holoprasinus* and *Palpomyia* species but at rates of 10 to 1,000 times the field use rates for mosquito control.

Some mortality has been observed against *Toxorhynchites* larvae that ingested prey immediately following intoxication. Filter feeding first instar *Toxorhynchites* larvae are as susceptible to Bti as mosquito larvae. Later instars are relatively insensitive to Bti when intoxicated prey are absent.

OTHER NON-TARGET SPECIES:

No toxicity was observed against crustacea including copopod species, Gambia, oysters, shrimp, crabs, mollusks, flatworms and amphibia.

In addition to safety to non-target vertebrates and invertebrates, Bti based larvicides have been used in large-scale mosquito and blackfly control programs for 20 years with no adverse effects on humans.

FORMULATIONS:

AQUABAC xt is an aqueous suspension specifically formulated to meet the needs of the mosquito control industry. The product is a tan, aqueous suspension. Formulation characteristics have been manipulated to suspend the active ingredient in the feeding zone of mosquito larvae for an extended period of time.

AQUABAC xt is a partially agglomerated aqueous suspension with a potency of 1,200 ITU/mg. Physical characteristics are: (1) viscosity; <1,200 cps, and (2) specific gravity; 1.06 to 1.10. These physical characteristics permit ULV ground and aerial application and also provide desirable suspension qualities when mixed with water. The formulation contains a high concentration of individual spores and protein crystals as well as agglomerated crystals. The agglomerates improve deposition when the formulation is applied ULV. The high number of individual spores and protein crystals enhances and prolongs the suspension of the product in the mosquito feeding zone.

AQUABAC xt APPLICATION INSTRUCTIONS

MOSQUITOES: HABITAT

RATE REQUIRED FOR CONTROL

Flood water, roadside ditches, irrigation ditches,
rice fields, pastures, woodland pools, snow melt ponds

0.25 to 0.50 pts/A
(300 – 600 ml/Ha)

Tidal water, salt marshes, catch basins,
storm water retention areas

0.5 to 1.0 pts/A
(600 – 1,200 ml/Ha)

Polluted water (sewage lagoons, etc.),
water with moderate organic matter,
water with a high concentration of suspended solids

1.0 to 2.0 pts/A
(1,200 – 2,400 ml/Ha)

AQUABAC xt may be applied in conventional aerial and ground application equipment with sufficient water to provide thorough coverage of the target area. The amount of water needed will be dependent on weather, type of spray equipment, and mosquito habitat.

Ground application should be made in 5 to 100 gallons per acre (50 – 1,000 liters per Ha) in conventional equipment. As low as one gallon per acre (10-lts/Ha) surface area can be used when the target is open with light vegetative cover. Aerial applications may be done diluted or undiluted. For undiluted applications, apply 0.25 – 2.0 pints per acre (300 – 2,400 ml/Ha) of **AQUABAC xt** through fixed wing aircraft or helicopters equipped with conventional boom and nozzles or rotary mist atomizers. For diluted applications, fill the mix tank or air craft hopper with the appropriate volume of water and agitate before adding **AQUABAC xt**. Maintain agitation during loading and spraying.

AQUABAC 200G is a granular formulation using corncob grit as the carrier. It is available in two mesh sizes, 5/8 and 10/14, and is effective against mosquitoes in a variety of habitats.

AQUABAC 200G APPLICATION INSTRUCTIONS

MOSQUITOES:

HABITAT

Flood water, roadside ditches, irrigation ditches, rice fields, pastures, woodland pools, snow melt ponds

Tidal water, salt marshes, catch basins, storm water retention areas

RATE REQUIRED FOR CONTROL *

2.5 to 10.0 lbs/A
(2.8–11.25 kgs/Ha)

*When late 3rd and early 4th instar larvae predominate, larval populations are high, or water is heavily polluted and/or algae is present, use 10 – 20 lbs/acre (11.25 – 22.5 kgs/Ha).

AQUABAC 200G should be applied uniformly in conventional aerial or ground equipment. A seven to fourteen day interval between applications should be employed. Longer periods of mosquito population suppression may result where sufficient numbers of non-target aquatic invertebrate parasites and predators are present, since these are not affected by **AQUABAC** and contribute to mosquito population reduction.

NUISANCE FLIES:

For control of the Nuisance Flies *Psychoda* spp. and *Chironomus* spp. With Aquabact xt in sewage treatment facilities utilizing trickling filter systems.

APPLICATION DIRECTIONS

<u>Nuisance Fly Habitat</u>	<u>Suggested Use</u>
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Rates ⁽¹⁾

Trickling filter system wastewater of wastewater treatment plants	10 – 50 mg per liter of feed to the system per 30 minutes.
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1. Use higher rates for control of *Chironomus* spp. Apply undiluted with a pre-calibrated pump or other device into the wastewater feeding into the filters for a minimum period of 30 minutes. Repeat applications as needed. Control of *Chironomus* spp. can take two weeks.

NUISANCE AQUATIC MIDGES:

For control of *Chironomine* midges (*Chironominae: Chironomini*) inhabiting shallow, man-made and natural lakes and ponds.

APPLICATION DIRECTIONS

<u>Nuisance Midge Habitat</u>	<u>Suggested Rate</u>
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Range ⁽¹⁾

Shallow lakes and ponds Per sewage oxidation ponds (less than an acre 6 feet deep)	1.0 gallon / acre (3.80 liters / acre)
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1. Apply diluted with water in a total volume of 5 gallons / acre by pouring or spraying over the surface to be treated with a pre-calibrated device. Repeat applications as necessary. Control of *Chironomini* midges can take up to two weeks.

FACTORS AFFECTING PERFORMANCE

Water conditions such as temperature, organic matter content, chlorine, some inorganic salts, and pH may affect performance of AQUABAC formulations. Larval feeding rates, subsequent ingestion of AQUABAC protein toxin and larval mortality rates are positively correlated with water temperature. Organic matter and algae represent an alternative food source for mosquito larvae. Therefore, the higher labeled rates are required a greater dose of AQUABAC for effective larval control.

Presence of free chlorine or some inorganic salts such as BaCO₃, K₂CO₃, and MgCO₃ can inhibit the activity of the protein toxin. The effectiveness of Bti is not impacted by pH levels near 7. Higher rates of AQUABAC are recommended for alkaline habitats with pH levels equal to or greater than 9.4.

The effectiveness of Bti is also influenced by larval feeding behavior. Lower mortality rates have been observed with all Bti based formulations among Anopheles larvae because of the genera's preference for feeding at the surface. Although AQUABAC xt has been specifically formulated to enhance its suspension in the upper feeding horizon, the protein toxin does gradually settle out of the upper levels of water becoming inaccessible to these larvae.

PERFORMANCE RESULTS

AQUABAC xt has been widely tested and used worldwide for the past fifteen years in mosquito control projects. It has been shown to be superior to other Bti products against Anopheline species in Indonesia, India, Malaysia, Singapore, and Taiwan.

Analysis of field results from trials conducted in the United States indicate 95 to 100% mortality against Culex, Aedes, and Anopheline species.

State	Mosquito Species	Type of Application	Rate/HA	% Reduction
LA	Psorophora columbiae	Diluted aerial	1,200 ml	100%
LA	Culex pipiens	Diluted ground	2,400 ml	97%
UT	Culex tarsalis	Diluted aerial	2,400 ml	95%
UT	Culex tarsalis	Diluted aerial	2,400 ml	98%
UT	Aedes darsalis Culex tarsalis Culex pipiens	Sand ground	2,400 ml	97%