# TECHNICAL USE BULLETIN





# WATER SOLUBLE POUCH







### History BACILLUS THURINGIENSIS SUBSP. ISRAELENSIS (STRAIN AM65-52)

*Bti* is a naturally occurring spore-forming bacterium found in soil and aquatic environments throughout the world. At the time of sporulation, *Bti* produces a highly specific delta-endotoxin, which is only toxic to larvae of mosquitoes, black flies, and closely related flies upon ingestion.

During 25 years of field use in a variety of settings around the globe, *Bti* has been shown to provide effective, reliable, and environmentally compatible control of mosquito larvae. In addition to the effectiveness of *Bti*, it has an excellent safety record and very low mammalian toxicity: LD50 values for both oral and dermal toxicity are more than 30,000 mg/kg. The mosquitocidal crystal proteins, spores, and vegetative cells of *Bti* administered by different routes have been found to be non-pathogenic and non-toxic to various animal species in maximum challenge tests.<sup>1</sup> *Bti* is safe for use in aquatic environments, including drinking water reservoirs, for the control of mosquito, black fly and nuisance insect larvae.<sup>2</sup>

### BACILLUS SPHAERICUS (STRAIN 2362)

*Bsph* is also a naturally occurring spore-forming bacterium found throughout the world in soil and aquatic environments. Early development of *Bsph* for mosquito control focused on strains isolated and maintained by the Pasteur Institute, WHO collaborating Center, Paris, France. Since 1995, *Bsph* has demonstrated the ability to provide residual control of mosquito larvae in a great variety of aquatic habitats. This biological larvicide is capable of providing residual control in highly organic environments, including catch basins, waste lagoons, animal waste ponds, septic tanks, irrigation ditches or roadside ditches.

#### **BIOFUSE™ TECHNOLOGY**

Microbial mosquito larvicides have gained considerable acceptance in recent years. The value of *Bti* and *Bsph* to mosquito control programs worldwide is well established.<sup>1</sup> *Bti* and *Bsph* each offer unique advantages relative to chemical insecticides. Both *Bti* and *Bsph* offer relative safety to humans and non-target organisms.<sup>2,3,4</sup> *Bti* provides broad spectrum activity, rapid control, and low potential for resistance; while *Bsph* exhibits extended residual control, efficacy in polluted water, and high target specificity.<sup>5</sup>

Due to these unique advantages, Valent BioSciences Corporation developed BioFuse<sup>™</sup> Technology, a proprietary (patent pending USA; patent approved EU) technology that combines *Bti* and *Bsph* in a specific toxin ratio into **every micro particle**. This technology offers mosquito control professionals the ability to take advantage of each microbial larvicide's strengths, while significantly reducing the limitations that each possesses.

#### **DEVELOPED "BY REQUEST"**

The water soluble pouch concept was developed at the request of mosquito control professionals. With the spread of West Nile virus in the United States beginning in 1999, Valent BioSciences Corporation was asked by mosquito abatement programs to develop a formulation that could be easily applied to catch basins for control of mosquito larvae. For this purpose, Valent BioSciences Corporation introduced VectoLex<sup>®</sup> WSP Biological Larvicide (active ingredient: *Bacillus sphaericus*) in 2001. Since then, the popularity of this product launched additional WSP formulations from other companies for both professional and consumer use.

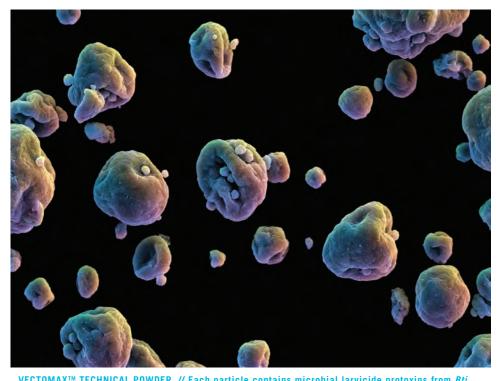
<sup>&</sup>lt;sup>1</sup> Becker N, Petric D, Zgomba M, Boase C, Dahl C, Lane J and A Kaiser. 2003. Mosquitoes and their control. Kluwer Academic; Plenum Publishers, New York: ISBN 0-306-47360-7

<sup>&</sup>lt;sup>2</sup> Lacey LA and RW Merritt. 2003. The safety of bacterial microbial agents used for black fly and mosquito control in aquatic environments. *In:* "Environmental Impacts of Microbial Insecticides: Need and Methods for Risk Assessment" (HMT Hokkanen and AE Hajek, eds.), pp 151-168. Kluwer Academic Publishers Dordrecht, The Netherlands.

<sup>&</sup>lt;sup>3</sup> World Health Organization, 1999, Environmental health criteria 217 microbial pest control agent *Bacillus thuringiensis*, WHO, Geneva Switzerland, ISBN 92 4 157217 5.

<sup>&</sup>lt;sup>4</sup> Siegel JP and DA Shadduk. 1990. Mammalian safety of Bacillus sphaericus, Chp. 21 in "Bacterial Control of Mosquitoes and Black Flies" (de Barjac & Sutherland eds.). Rutgers University Press, ISBN 0-8135-1546-7

<sup>&</sup>lt;sup>5</sup> Lacey LA. 1990. Chp. 18 In: "Bacterial control of mosquitoes and black flies" (deBarjac H and DJ Sutherland eds.). Rutgers University Press, New Brunswick, NJ.



VECTOMAX<sup>TM</sup> TECHNICAL POWDER // Each particle contains microbial larvicide protoxins from *Bti* and *Bacillus sphaericus*.

VectoMax<sup>®</sup> WSP Biological Larvicide is an advanced biological mosquito larvicide based on Valent BioSciences Corporation's new proprietary BioFuse<sup>™</sup> Technology, which combines the time-proven and environmentally compatible microbial active ingredients *Bacillus thuringiensis* subsp. *israelensis (Bti)* and *Bacillus sphaericus (Bsph)* **into a single micro particle**. Thus, mosquito larvae get a dose of a carefully selected ratio of both toxins when VectoMax<sup>®</sup> particles are ingested.

VectoMax<sup>®</sup> WSP is a Water Soluble Pouch containing VectoMax<sup>®</sup> CG. The formulation is ideal for treatment of catch basins and other small breeding sites. The pouch is placed directly into the breeding site and is very useful for control of West Nile virus (WNV) vectors in urban and suburban areas.

#### FEATURES

- Quickly kills mosquito larvae
- Controls all mosquito species
- · Highly specific activity on mosquitoes
- · Can be used in clean and polluted habitats
- Offers residual control of several mosquito species
- Up to 8 weeks of residual control under typical environmental conditions
- Combination microbial
- Water soluble packaging

#### **BENEFITS**

- Results assessed quickly in the field
- Not harmful to non-target organisms
- Reduced number of applications
- Dust-free
- Easy to apply (malleable, slips into tight spots)
- Does not get hung up in debris or buried in sludge at the bottom of a catch basin
- Clean to the touch

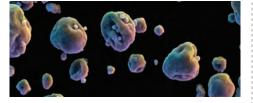


### Mode of Action

Both *Bti* and *Bsph* produce complex crystal proteins known as protoxins during sporulation. When these proteins are applied to larval habitats of mosquitoes, the mosquito larvae ingest them by filter feeding. The crystal proteins are solubilized by the alkaline juices in the larval midgut and are cleaved by the midgut proteases, yielding active peptide toxins called delta-endotoxins. The delta-endotoxins cause the formation of holes in the midgut cell wall, leading to lysis of cells and larvae death.

However, activity of the delta-endotoxin for *Bsph* differs from that of *Bti* in several important ways. For *Bsph*, the toxin is attached to the bacterial spore, while *Bti* toxins are not attached to the spore (parasporal). The toxins of *Bsph* and *Bti* bind to chemically different receptor sites on cells. They are not related immunologically and are thought to have completely different molecular modes of action.

Operationally, the most important differences between the toxins of Bsph and Bti are speed of action and persistence in natural larval habitats. Bsph toxin is much slower-acting than Bti toxin. Larval mortality can take several days but is usually expressed within 48 hours of ingestion, while Bti provides quick kill. Initial results can be seen within 2-24 hours. Bsph toxin is also much more persistent in natural larval habitats than Bti. This persistence is thought to be the result of a combination of features, including protection of the protein by the spore coat; slower settling rate; and the unique ability of Bsph spores to germinate, grow, and produce toxins in cadavers of mosquito larvae treated with the material. VectoMax® WSP combines the quick kill seen with Bti in combination with the residual properties of Bsph.



### Application

Each 10-gram packet of VectoMax<sup>®</sup> WSP treats up to 50 square feet of mosquito larval habitat. Once placed in water, VectoMax<sup>®</sup> WSP dissolves in 2-5 minutes, releasing the VectoMax<sup>®</sup> granules. The carrier disperses on the water surface and in the water column, distributing micro particles containing both *Bti* and *Bsph* into the larval feeding zone.

### **APPLICATION RATES**

For control of mosquito larvae in the following sites:

HABITAT	RATE/RANGE	
Drainage/Drainage Systems: 1		
Storm drains, catch basins,	1 pouch/50 sq. ft.	
retention, detention, and		
seepage ponds.		
Treatment Areas (For Use In): <sup>2</sup>		
Ponds	Flowerpots and planters	
Lagoons	Snowmelt pools	
Water gardens	Septic tanks	
Hollow trees and tree holes	Abandoned swimming pools	
Urns	Unused swimming pools or spas	
Rain barrels	Flooded basements	
Livestock watering troughs/	Pool covers	
ponds/tanks	Gutters and drains	
Irrigation ditches	Wheelbarrows	
Roadside ditches	Garbage cans and covers	
Impounded wastewater	Discarded tires	
associated with fruit and	Any location where water	
vegetable processing	accumulates and remains	
Animal waste lagoons	standing for periods of time	
Flood water	except treated, finished	
Standing water	drinking water for human	
Birdbaths	consumption	
Fountains		

Treat on basis of surface area of potential mosquito breeding sites by placing one (1) VectoMax<sup>®</sup> Water Soluble Pouch for up to 50 square feet of treatment area. Re-apply as needed (up to 8 weeks under typical environmental conditions).

<sup>&</sup>lt;sup>2</sup> Treat on basis of surface area of potential mosquito breeding sites by placing one (1) VectoMax<sup>®</sup> Water Soluble Pouch for up to 50 square feet of treatment area. Re-apply as needed (up to 4 weeks under typical environmental conditions). 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 the product and contribute to mosquito population reduction.

## Storage and Disposal

Once the foil bag containing Water Soluble Pouches is opened, minimize exposure of WSP to humidity.

Do not contaminate water, food, or feed by storage or disposal.

**Pesticide Storage:** Store in a cool, dry place. **Pesticide Disposal:** To avoid wastes, use all material in this container by application according to label directions. If wastes cannot be avoided, offer remaining product to a waste disposal facility or pesticide disposal program. (Often such programs are run by state or local governments or by industry.)

**Container Disposal:** Non refillable container. Do not reuse or refill this container. As long as WSP is unbroken, dispose of the empty outer foil pouch in the trash or by other procedures approved by state and local authorities, or offer for recycling, if available.

### Effects on Non-Target and Beneficial Organisms

Both *Bti* and *Bsph* have excellent safety records and very low mammalian toxicity. Effects of label-approved applications on non-target organisms are practically nonexistent.<sup>1</sup>

DRGANISM	STUDY TYPE	RESULT
Odonata		
Dragonflies/Damselflies		
T. corruptum	Lab/naiads fed infected larvae	No effect
E. civile	Lab/naiads fed infected larvae	No effect
Ephemoptera		
Mayflies		
C. pacificus	Field treatment ( <i>Bti</i> technical powder 0.56 kg/ha)	No effect
C. pacificus	Field treatment ( <i>Bsph</i> technical powder 0.22 kg/ha)	No effect
Heteroptera		
Corixids/Notonectids		
C. decolor	Field treatment ( <i>Bti</i> technical powder 0.56 kg/ha;	No effect
	Bsph technical powder 0.25 kg/ha)	
N. undulata	Lab/fed infected larvae	No effect
A. bouvieri	Lab/LC50 (Bsph)	500X mosquito LC50
N. undulata	Lab/fed infected larvae	No effect
N. unifasciata	Field study/treated ponds	No effect
Buenoa sp.	Field study/treated ponds	No effect
Coleoptera		
Dytiscidae	Field studies	No effect
Hydrophylidae	Field studies	No effect
Crustacea		
Daphnia		
E. bampo	Laboratory ( <i>Bti</i> )	100-200X mosquito rate
D. similis	Laboratory (Bsph)	Effect at 27,000X mosquito rate
Fairy Shrimp		
S. dichotomus	Laboratory (Bsph)	Effect at 15,000X mosquito rate
Crawfish		
P. clarkii	Laboratory	Effect at 1,000X mosquito rate

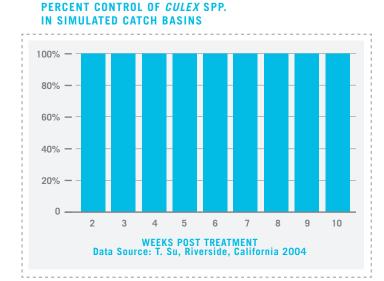
<sup>1</sup> Lacey and Mulla (1990). Safety of *Bacillus thuringiensis* subsp. *israelensis* and *Bacillus sphaericus* to non-target organisms in the aquatic environment. *In* "Safety of Microbial Insecticides" (Marshall Laird, Lawrence Lacey, and Elizabeth Davidson eds.), Chap. 12. CRC Press, Inc. Boca Raton, Florida. (Unless otherwise indicated, studies include evaluations of both *Bti* and *Bsph*.)

Bti and Bsph have been extensively tested, and they are not human health hazards when handled as instructed by the product label. VectoMax<sup>®</sup> WSP has a hazard classification signal word of *Caution*.

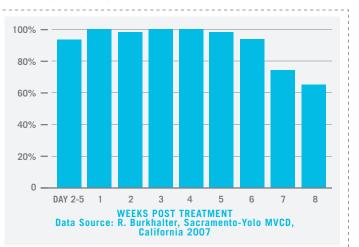
# Results You Can See

Based upon extensive field evaluations, VectoMax<sup>®</sup> WSP persists for up to 8 weeks following a single application under typical environmental conditions. In addition, VectoMax<sup>®</sup> WSP provides quick kill of all mosquito species in both clean and polluted waters while offering the ability to quickly assess results in the field.

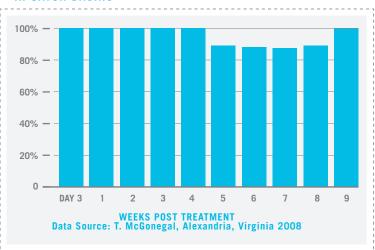
Duration of residual control is generally determined by habitat factors rather than by application rate. Factors that can reduce residual activity include flushing and water movement, water depth, and possibly chemical contamination.



PERCENT CONTROL OF *CULEX* SPP. IN CATCH BASINS



### PERCENT CONTROL of *CULEX* SPP. IN CATCH BASINS



VECTOMAX, VECTOLEX, BIOFUSE and VALENT BIOSCIENCES are registered and/or trademarks of Valent BioSciences Corporation. Valent BioSciences Corporation owns registrations for these marks in the United States and elsewhere.



870 Technology Way / Libertyville, IL 60048 © Valent BioSciences Corporation March 2009

AG 5404

