VectoBac®

Technical Use Bulletin for ***VectoBac 12AS***
Mosquito and Black Fly Larvicide

VALENT BIOSCIENCES CORPORATION




VectoBac® 12AS is an aqueous suspension formulation of Bacillus thuringiensis for control of mosquito and black fly larvae. VectoBac 12AS has a potency of 1200 ITU's per milligram equivalent to 4.84 billion ITU's per gallon (1.28 billion ITU's per liter).

VectoBac viscosity is less than 500 CPS at 25°C (77°F). VectoBac 12AS is light brown in color with a specific gravity slightly greater than 1.0. VectoBac aqueous suspension is produced directly from fermentation slurry which helps insure an average particle size of 2 to 9 microns. The small particle size improves efficacy by maintaining maximum availability of particles in the feeding zone for mosquito and black fly larvae. Small particles stay suspended for a longer period of time in the target larval feeding zone.

VectoBac 12AS is available in the following package sizes:

- 2.5 gallon (10 liter) plastic jugs, packaged 2 per case
- 50 liter plastic drums (international only)
- 30 gallon plastic drums
- Bulk shipments (4,000 to 5,000 gallon tanker trucks, U.S. only)

Mixing Instructions

VectoBac 12AS should be shaken well or agitated before use and then added to the desired quantity of water in the mix tank during agitation. The resulting suspension resists settling. However, mild agitation should be maintained during spraying to assure a uniform mix. Do not mix more VectoBac 12AS with water than will be used in a 2 day period.

Mosquito Control Applications

Ground

VectoBac may be applied either diluted in water or in undiluted ULV applications.

For diluted applications, apply desired amount of VectoBac 12AS in 5 to 100 gallons of water per acre (50 to 1000 liters per hectare). Applications may be made with hand pump backpack sprayers, compressed air sprayers, air blast units, mist blower sprayers, power backpack sprayers or hydraulic sprayers. VectoBac 12AS may also be applied with direct injection systems where this application technique is preferred. Calibration of spray unit to assure proper delivery rates, and an application technique that assures even coverage of area treated are essential to success. For a detailed discussion of calibration methods, consult "Successfully Calibrating Applications of VectoBac and VectoLex" VBC Brochure, May 2000 AG5293.

Ground ULV applications can successfully be made using truck mounted, backpack or hand held equipment. These applications are dependent upon proper wind and atmospheric conditions for successful coverage of the breeding site. Applications using truck mounted equipment may be impaired by obstructions such as buildings and dense vegetation. Backpack and/or hand held equipment may be of greater utility when making ground ULV applications in urbanized or densely vegetated areas, because of increased access to the breeding areas. Diluted high volume applications may be effective in a broader range of conditions. Droplet size recommended for ground ULV applications is 20um-100um. Smaller droplets will produce larger swaths due to down-wind movement, and may penetrate vegetation and certain micro-habitats more efficiently. Larger drops will provide greater surface deposit and more consistent deposition in narrower swaths.

Aerial

VectoBac 12AS may be applied either undiluted or diluted with water. For undiluted applications, apply VectoBac 12AS at 0.25 to 2.0 pints per acre (0.3 to 2.5 liters per hectare), by fixed wing or helicopter aircraft equipped with either conventional boom and nozzle systems or rotary atomizers. Avoid excessive recirculation during application of undiluted VectoBac 12AS as this may change physical properties.

For diluted applications, the appropriate rate of VectoBac 12AS should be mixed with the desired amount of water. Maintain mechanical or hydraulic agitation to provide moderate circulation while loading or spraying.

Selection of application volume/acre and droplet size range will depend on site characteristics, atmospheric conditions and local experience. For sites having very uniform, low vegetative cover such as rice fields, fine sprays in 50-100 UM micron VMD droplet diameter range and spray volumes as low as 4 ounces per acre (0.3 liters per hectare) have been effective. However, aerial applications using droplets in this size range are substantially affected by atmospheric conditions. Considerable downwind movement (drift) should be accounted for. Such applications should not be made if wind exceeds 5 mph or thermal activity is present. Arid conditions may result in significant evaporative loss of small droplets.

For these reasons, a larger droplet size and higher volumes are generally required. Consistent results can be achieved with total spray volumes of 16 oz to 5 gallons per acre. Droplets in the 100-1000um range provide more consistent ground deposit than smaller drops. Volume of ground deposit will increase with droplet size. However, coverage (drops/unit area) will decrease, and swath width may decrease. The ideal droplet size range for local conditions will depend upon habitat and atmospheric conditions. The addition of spray adjuvants that retard evaporation, and enhance foliage penetration may also improve consistency of results.

A variety of aerial spray systems can be adapted to achieve the appropriate droplet size and coverage. When spray volumes >1 gpa are employed, DC, CP or Flat fan nozzles have all been successful. A zero degree of "straight back" nozzle orientation will produce larger droplets than a downward deflection of the nozzle. Deflection should not exceed 45°. At least 20 nozzles should be used across the boom, which should not exceed 75% wingspan.

Aerial application equipment should be characterized to verify swath width, droplet spectrum and distribution. Optimum applications will result when atmospheric conditions and characterization data are factored into the selection of lane separation, altitude and treatment offset. Atmospheric conditions can significantly impact success of aerial applications. High winds (>10mph), low humidity, and unstable atmospheric conditions negatively affect results.

Mosquito Control Applications

Habitat	*VectoBac 12AS*
Irrigation ditches, roadside ditches, flood water, standing ponds, woodland pools, snow melt pools, pastures, catch basins, storm water retention areas, tidal water, salt marshes, and rice fields	0.25 to 1 pints per acre (0.3 to 1.2 liters per hectare)
Polluted or highly organic water* (such as sewage lagoons and animal waste lagoons)	1 to 2 pints per acre (1.2 to 2.4 liters per hectare)

**Use higher rates in polluted water and/or when 3rd and early 4th instar larvae predominate, mosquito populations are high, and/or algae are abundant. NOTE: Where mosquito populations, particularly in salt marsh conditions, exceed 40-50 larvae per dip, higher rates of VectoBac 12AS should be used for more effective control.*

Bulk Handling (Tanker Handling)

Upon arrival, the contents of the tanker should be recirculated once before unloading for use. This can best be achieved by pumping the material from the outlets value back through the open manhole, or, if possible from end to end. Use a pump with sufficient capacity, for example, a 5 horsepower gasoline engine powered centrifugal pump or a positive displacement gear pump capable of moving at least 100 GMP (6.3 liters per second). The manhole of the tanker must always be open when pumping to prevent the collapse of the tanker walls. When unloading the tanker into a holding tank, the tank and lines should be cleaned by thoroughly rinsing with water.

If the tanker is being spotted, it must be placed on solid ground with the front support on oak boards. If unloaded into a compartmentalized tanker, always unload into the middle compartment first, the wheel end and then the dolly end. Reverse the procedure when unloading a compartmentalized tanker. This procedure will prevent tipping the tanker.

During storage, material should be recirculated at least once every 14 days to prevent separation, and it should be recirculated prior to use if it has been sitting for 4 days or more.

Black Fly Control Applications

Where to Apply

Black flies can be found breeding wherever there is running water. Although some species can be found in merely a trickle of water, most prefer rapid water flows high in oxygen content. When mapping treatment sites, look first in the rapids, waterfalls, and dam outlets. It is advised to map these areas under different flow conditions as the breeding sites will change during the course of a season.

When to Apply

The timing of the treatments is probably the most critical factor in effective black fly control. It is desirable to begin treatment before the adult black flies become a problem! Since VectoBac kills only black fly larvae, not the eggs or pupae, the treatments should be timed when the majority of the population is still in the larval stages, preferably just before the insect enters the final larval stage. Final instar larvae can be recognized by predominate black spots, the respiratory histoblasts, on either side of the thorax.

It is advisable that the life history of the black fly species being treated be determined before major efforts in control are initiated. The water temperature is a good indicator of larval development for most black fly species and can be used to determine approximate treatment initiation and treatment cycles. At water temperatures of 78°F (25.5°C), development of black fly larvae can take less than 7 days, whereas at 50°F (10°C), development may take over 30 days. It should be noted that water temperatures will vary over the length of a river, therefore, monitoring should be done carefully at many sites in the river/stream to be treated.

How to Apply

VectoBac 12AS can be applied undiluted or diluted using aircraft, hand held sprayers, watering cans or directly from the 12AS containers, depending upon the type and size of the target river/stream. Small streams are usually treated by hand, whereas, large rivers and streams are treated by boats or aircraft.

Treatments should be made far enough upstream of the larval attachment sites to allow ample time for the VectoBac 12AS to mix throughout the water column and provide ample time for the larvae to feed. The exact distance that material is injected into the water above the larval attachment sites will depend upon many factors. Ideally, VectoBac should move in a well dispersed "slug" through the attachment sites for a period of 10 minutes or longer. To accomplish this, treatments need to be made upstream for at least 50 ft (15 M) in small rivers/streams and as much as 150 ft (46 M) in large river systems. Rate of application will be determined by the stream discharge and the required amount of VectoBac necessary to achieve a 0.5 to 25 ppm concentration in the river/stream water. Under normal river conditions, 1.2 ppm or 10 minutes (equal to 720 ml/1000 1/sec or 1 m3/sec discharge) will control most black fly species, most of the time. When water temperatures are less than 50°F (10°C) and under high silt loads or concentrations of planktonic algae, the rate should be increased to 2.5 ppm (1500 ml/1000 1/sec.) Treatment should be avoided in water clogged with vegetation and above deep pools. Care should be taken to treat the entire width of the river/stream, as many larvae can be found along the banks.

Troubleshooting Control Problems

Excellent control with any *Bti* product depends upon qualified applicators and control personnel with a good understanding of the complex relationships of black fly biology and the environment in which black flies inhabit. Black fly mortality depends upon several factors including the amount of VectoBac the larvae are able to trap and ingest, the duration of the exposure, the concentration of the exposure, and the rate of feeding. Feeding rate depends upon larval stage, concentration of suspended particles, temperature of the water, particle size of the Bti, and black fly species.

Following are a few examples of operational conditions that may effect black fly suppression programs:

Condition	*Possible Solution*
◆ Water contained high (>1500 cells/ml) of planktonic algae or other suspended solids.	Increase VectoBac Rate
◆ Larvae feed slowly, don't receive lethal dose of *Bti*; Water temperature too cold (<10°C)	Increase VectoBac Rate
◆ VectoBac passed over larvae too fast to allow enough time to ingest lethal dose of *Bti*.	Application too close to attachment site, move application further upstream
◆ VectoBac settles out before reaching attachment	Application was too far upstream; Check site for deep pools or aquatic vegetation between application site and attachment site
◆ Poor or erratic control	Time in applying VectoBac may be too long; Bti became too diluted to be effective to all larvae at the attachment site. Increase exposure time.
	River discharge was underestimated; dosage was too diluted to give good control. Check discharge measurement calculations for accuracy.

A successful black fly control program requires standard procedures. The following methods are provided as guidelines for proper ground and aerial application of VectoBac liquid formulations in streams and rivers. Since the quantity of VectoBac required to achieve a desired dosage is dependent primarily on stream flow, it is necessary to calculate the discharge of a stream immediately preceding the point of application. This measurement should be made in a section of the stream which is straight and free-flowing. Pools, bends, turbulence, wind gusts, and debris may cause inaccurate results.

Calculations Prior to Treatment

1. Stream Cross Sectional Area (square meters)

Measure stream width at the application site. Measure depth at 5 to 10 points at equal intervals across the stream width. Calculate average depth of stream. Multiply depth by width to determine stream cross sectional area (Depth x Width = stream cross sectional area in square meters).

2. Flow Velocity (meters/second)

Use citrus peel, small stick, or other buoyant object to measure flow rate of stream. Mark off 10 meter length of stream. Use a stopwatch to determine the length of time required for the buoyant object to travel the 10 meters. Divide the 10 meters by the time measurements (seconds) to determine meters per second. Repeat process three times and take an average of the meter per second tabulations.

3. Flow Volume (cubic meters/second)

The flow volume is equal to the stream cross sectional area multiplied by the flow velocity. This number represents the amount of water that will pass through a distance during a measured period of time. (Stream cross sectional area x flow velocity = flow volume in cubic meters/second.)

4. VectoBac Required For Treatment

Flow volume x 1,000 x 60 = liters/minute of stream flow Liters/minute x 1,000 = ml/minute

$$\text{ml of VectoBac} = \frac{\text{desired rate (ppm) x ml/minute}}{1{,}000{,}000}$$

5. Application of VectoBac

Apply required amount of VectoBac to stream over a period of 1 to 15 minutes by slowly applying product across stream width. If quantity to be applied is a very small amount (less than 4 liters) consider dilution with water prior to treatment to increase volume.

6. Suggested Rates and Application Guidelines

- 0.05 to 2.5 ppm for 10 minutes
- 0.5 to 25 ppm for 1 minute
- Swift streams (500 - 1000 m^3/second) generally require shorter dosage times.
- Polluted or silty water generally requires higher rates.
- Cold water (7°C to 1O°C or 45°F to 50°F) temperature requires higher rates.
- Rough bottom streams require higher rates
- Good dispersion of VectoBac improves control

NOTE: Each river system is unique in its stream bed morphological/substrate profile, water chemistry and hydrological conditions. Stream and river dynamics are influenced by a gradient of physical factors formed by the drainage network. Black fly larval distribution is determined by hydrodynamics created by the uniqueness of the river/stream system. It is impossible to make specific recommendations on dose level and injection time that will provide effective control in all streams.Therefore, breeding sites should be properly identified and mapped, and pilot studies conducted to ascertain correct downstream distance of larval control (carry).

Conversion Factors:

Cubic ft. x 7.48 = gallons
Cubic ft./min. x 0. 1 247 = gals./sec.
Cubic meters x 1000 = liters
Cubic meters x 264.2 = gallons
Feet/min. x 0.3048 = meters/min.
Feet/sec. x 18.22 = meters/min.
Milliliters x 0.001 = liters

Ounces (fluid) x 0.0296 = liters
Square miles x 640 = acres
Area of a Rectangle = length x width
1 gal. = 3.7854 liters = 4 quarts
1 lb. = 0.4536 kg. = 453.54 gms. = 16 oz.
1 qt. = 0.9464 liter

Field Rates of VectoBac 12AS per Unit Stream Flow for Black Fly Control

Concentration In PPM	U.S. Measure OZ. Product Per 1000 Cubic feet	Metric Measure ML Product per Cubic Meter
1	1	1
5	5	5
10	10	10
15	15	15
20	20	20
25	25	25
30	30	30
40	40	40
50	50	50

Tank Mix Stability

Tests have shown that the VectoBac 12AS formulation will maintain useful activity for a period of 2 days when mixed with normal tap water. Diluted VectoBac tank mixes should be recirculated prior to use to maintain uniform suspension.

Storage

VectoBac aqueous suspension should be stored in cool locations and out of direct sunlight. At storage temperatures up to 24°C or 75°F, useful life is expected to be maintained for a minimum of 2 seasons. In northern areas, product can be held in unheated storage facilities. Freezing will not reduce potency; however, the product must be thoroughly mixed after thawing and prior to use.

Pesticide Disposal

Wastes resulting from the use of this product may be disposed of on site or at an approved waste disposal facility.

Container Disposal

Triple rinse (or equivalent). Then puncture and dispose in a sanitary landfill, or by incineration, or, if allowable by State and local authorities, by burning. If burned, stay out of the smoke. Do not reuse the container. Do not contaminate potable water, food or feed by storage or disposal.

Precautions: VectoBac aqueous suspension can cause corrosive effects to pure aluminum spray system components if diluted or undiluted product is left in prolonged contact with these parts. Rinse spray system with plenty of clean water after each use.

Avoid spraying undiluted product and product mixtures over automobiles as the dried deposits may be difficult to wash off.

Results of Mosquito Trials with VectoBac 12AS

Mosquito Species	Habitat	Investigator	Method of Application	Rate Pts/A (L/Ha)	Larvae Reduction
Ae. Vexans	Flood water	Dr. Gary L. Benzon Cape May County, NJ	Ground	0.5 (0.6)	98-100% 48 hrs.
Ochlerotatus taeniorhynchus	Ponds	Dr. Geoffrey Scott U of S.C. McClellanville, SC	Ground	0.5 (0.6)	100% 24 hrs.
Psorophora Columbiae	Rice	Dr. Max Meisch Fayetteville, AR	Ground	0.5 (0.6)	100% 24 hrs.
Culiseta inornata	Brackish	Dr. Richard Garcia Alameda County, CA	Ground	0.5 (0.6)	96% 48 hrs.
Culex tarsalis *Anopheles freeborni*	Rice fields	Dr. Richard Garcia Yuba County, CA	Air	2.3 oz (0.15)	96% 48 hrs.
Ochlerotatus sollicitans	Salt marsh	Mr. Cy Lesser Somerset County, MD	Ground	0.5-1.0 (0.6-1.2)	96% 48 hrs.
Ochlerotatus sollicitans	Salt marsh	Mr. Cy Lesser Dorchester County, MD	Air	0.5-1.0 (0.6-1.2)	95-98% 48 hrs.
Ochlerotatus taeniorhynchus	Mangrove	Dr. R. Parsons Sarasota County, MCD Marathon, FL	Air	2.0 (2.4)	98-100% 24 hrs.
Ochlerotatus	Pasture	Allan Inman	Air-ULV	1.5 (1.8)	100%

Results of Black Fly Trials with VectoBac 12AS

Black Fly Species	Habitat/Location	Investigator	Method of Application	Rate ppm/min	Mean Larval Mortality	Carry
Simulium jenningsi	Susquehanna River, Allegheny River Delaware River	Dan Arbegast PA DEP Harrisburg, PA	Aerial Aerial Aerial	11.5 11.5 11.5	95% 90% 98%	0.25-2 mi 1 mi 1 mi
S. jenningsi	Yellow Breeches Creek	Doug Orr PA DEP Harrisburg, PA	Ground	10	95%	1.5-2.5 mi
S. confusum *S. tuberosum*	Duncan Creek	Elmer Gray Clemson Univ. Clemson, SC	Ground	25	93%	2.25 mi
S. jenningsi	Tyger River	Elmer Gray	Ground	10	91%	5.8 mi
S. jenningsi	New River	Janice Smithson WV Dep, St. Albans, WV	Aerial	11	90%	0.5-1 mi
S. vittatum	Jordan River	Ken Minson Salt Lake City, UT	Ground	25	93%	0.5-1 mi

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