

*Suffolk County Vector Control & Wetlands
Management Long Term Plan
& Environmental Impact Statement*

**TASK 12: EARLY ACTION PROJECTS
CAGED FISH EXPERIMENT**

PESTICIDES EFFICACY TESTS

Submitted to:

**Suffolk County Department of Public Works
Suffolk County Department of Health Services
Suffolk County, New York**

Submitted by:

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**SUFFOLK COUNTY VECTOR CONTROL AND WETLANDS MANAGEMENT
LONG - TERM PLAN AND ENVIRONMENTAL IMPACT STATEMENT**

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LIST OF ABBREVIATIONS AND ACRONYMS

ABDL	Arthropod-Borne Disease Laboratory
<i>Bti</i>	<i>Bacillus thuringiensis israelensis</i>
CA	Cashin Associates, P.C.
EPA	Environmental Protection Agency
NYSDEC	New York State Department of Environmental Conservation
SCVC	Suffolk County Vector Control
ULV	Ultra Low Volume

1. Introduction

As part of the Caged Fish Study approved by New York State Department of Environmental Conservation (NYSDEC), DEC permit # 1-4700-00007/00037, tests were conducted to ascertain if pesticides from Suffolk County Vector Control's (SCVC) aerial operational sprays were reaching the caged organisms. Caged adult *Culex pipiens* mosquitoes were used to test the efficacy of Scourge® 18-54 (resmethrin). When the larvicide Altosid® (20 percent S-methoprene) was used during aerial operational sprays, the emergence of the mosquito larvae into adults was assessed.

2. Scourge® 18-54 (Resmethrin)

2.1 Background

Scourge® is a mosquito control adulticide used by SCVC since 1995. Its active ingredient is resmethrin, which has been registered by the Environmental Protection Agency (EPA) for pesticide use since 1971. The formulation of this adulticide consists of 18 percent resmethrin and 54 percent piperonyl butoxide and in this study was applied by a helicopter using an ultra low volume (ULV) spray.

The purpose of this study is to assess whether or not this adulticide is reaching the locations of the cages housing the aquatic organisms, sheepshead minnow (*Cyprinodon variegatus*) and grass shrimp (*Palaemonetes pugio*) by determining the mortality rate of the caged adult mosquitoes after the spray event.

2.2 Materials and Methods

Scourge® was used in two ULV aerial applications on August 18, 2004, and August 25, 2004, following normal SCVC procedures. The application rate of its active ingredient, resmethrin, is 0.6 oz/acre.

One pair of marsh sites was used: Johns Neck, located in Mastic, as the field spray site and Havens Point, located in East Moriches, as the control or reference site.

Test organisms used are female *Culex pipiens* from laboratory colonies maintained in the Suffolk County Department of Health Service's Arthropod-Borne Disease Laboratory (ABDL).

Nets for the caged mosquitoes are six inches in diameter and two inches deep with a mesh screen on both circular surfaces, affixed with a rubber band one-half inch from the outer surface. At each marsh site, three nets were affixed vertically on stakes and placed in a row in close proximity to the caged organisms. The three stakes were approximately 33 inches or 2.75 feet apart from each other in a row. The height of the nets was approximately 56 inches or 4.6 feet above the ground (Figure 1). When deployed, each net was checked to ensure that there were 10 live mosquitoes per net. Retrieval of the mosquitoes occurred at least a half hour after the operational spray is completed.

2.3 Results

August 18 Adulticide Spray at Johns Neck with Havens Point as a reference site

The mosquito nets were deployed at Johns Neck at approximately 7:15 p.m. on August 18, 2004. It took approximately 10 minutes to set up the three nets. The nets were deployed at Havens Point at approximately 7:50 p.m. The helicopter spray at Johns Neck took place at approximately 8:15 p.m. The nets at Johns Neck were retrieved at approximately 9:05 p.m. and the state of the mosquitoes was noted. At approximately 9:50 p.m. the nets were retrieved at Havens Point, after which time all the nets were returned to the ABDL. All the mosquitoes in the 3 nets at Johns Neck were dead upon retrieval of the nets while all the mosquitoes from the nets at Havens Point were alive. Refer to Table 1, which lists the results from both aerial adulticide spray events.

8/25 Adulticide Spray at Johns Neck with Havens Point as a reference site

The cages of the test aquatic organisms for the second adulticide event on August 25, 2004, were placed in the ditches on the same day, August 25, 2004 as the spray event with no larvicide event occurring on or near this day. An additional caged site at the end of a ditch at Johns Neck marsh and Unchachogue Creek was deployed; this location was close to the placement of the mosquito nets. At Johns Neck, the mosquitoes were deployed at approximately 7:15 p.m. Nearby, a fourth mosquito net under the canopy was deployed. The operational spray was completed at approximately 8:30 p.m. and the nets were retrieved at approximately 9:10 p.m. At Havens Point, the nets were deployed at approximately 8:10 p.m. and retrieved at 9:55 p.m. after which time all the nets were returned to the ABDL. Three hours after the operational spray was

completed, the mortality rate at Johns Neck was 93.3 percent for the mosquitoes near the caged organisms versus a 13.3 percent mortality rate at Havens Point.

2.4 Discussion

The high mortality of the adult caged mosquitoes at the field spray site at Johns Neck indicates that the aerial applications of Scourge® did reach the caged organisms. The results of the mosquito net placed under the canopy at Johns Neck suggests that the canopy impeded the delivery of Scourge® to these mosquitoes.

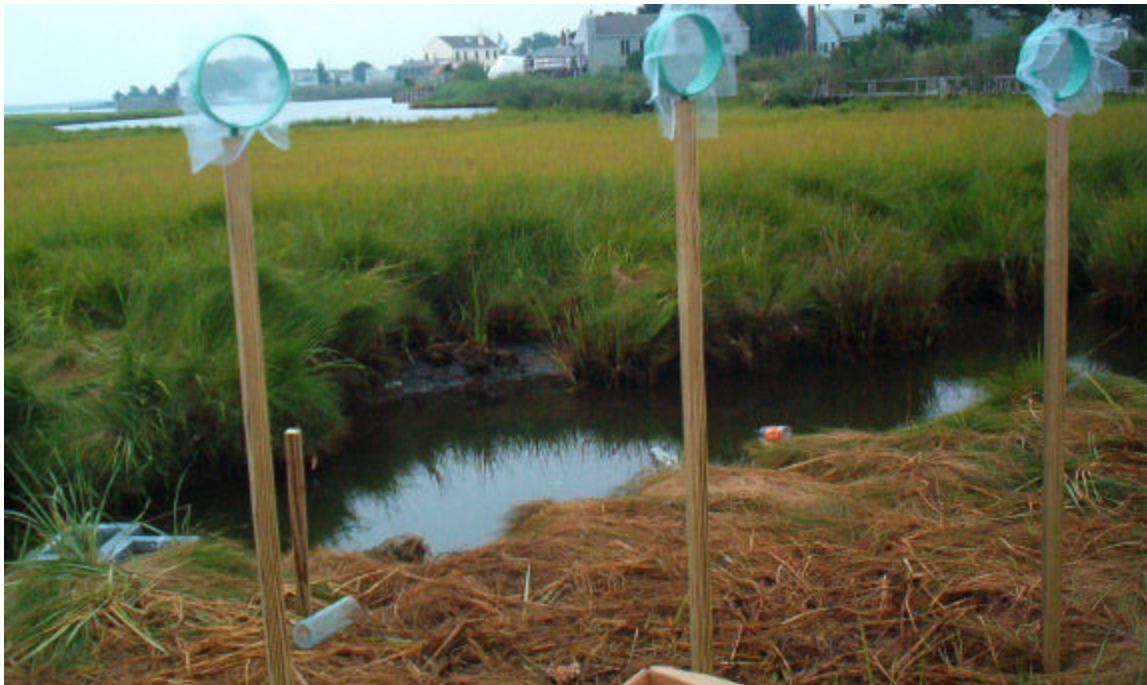


Figure 1 - Johns Neck Creek. Three mosquito nets near caged aquatic organisms . August 18, 2004 at approximately 6:15 p.m.

Table 1 – Caged Mosquitoes

Date	Net #	Number of adult mosquitoes alive 2 hours after spray exposure		% Mortality	
		JN	HP	JN	HP
8/18/04	1	0	10	100%	0%
	2	0	10	100%	0%
	3	0	10	100%	0%
Date	Net #	Number of adult mosquitoes alive 2 hours after spray exposure		% Mortality	
		JN	HP	JN	HP
8/25/04	1	3	9	70%	10%
	2	5	9	50%	10%
	3	4	10	60%	0%
	4*	10	4th mosquito net not used	0%	0%
Date	Net #	Number of adult mosquitoes alive 3 hours after spray exposure		% Mortality	
		JN	HP	JN	HP
8/25/04	1	1	9	90%	10%
	2	0	7	100%	30%
	3	1	10	90%	0%
	4*	10	4th mosquito net not used	0%	

* Mosquito net placed under canopy

Key: **JN** - Johns Neck in Mastic, Spray site

HP - Havens Point in East Moriches, control site that was not sprayed

3. Altosid® (20 percent S-methoprene)

3.1 Background

Altosid® has been used as part of SCVC’s larvicide program since 1995. The formulation used for its aerial helicopter ULV spray contains 20 percent (wt./wt.) s-methoprene. S-methoprene was registered for use by the EPA as a pesticide since 1985. This part of the study attempts to ascertain whether methoprene made its way to the salt marshes where the aquatic organisms were held, by examining for the emergence of larvae into adults. Methoprene does not cause immediate death of mosquito larvae, but rather it prevents emergence of adults from the pupae. This means that, in order to determine if a methoprene application is effective, larvae or pupae must be observed through their development to see if emergence is inhibited. Methoprene is sometimes used in combination with *Bacillus thuringiensis israelensis (Bti)*, a bacterial product that has been in use in Suffolk since 1982. Unlike methoprene, *Bti* does kill the larvae within 24 hours, which means its effects can be observed rapidly. When these two materials are applied in combination, *Bti* will normally kill the larvae before the effects of methoprene can be observed.

3.2 Materials and Methods

Two sites were chosen for the larval studies:

- Timber Point marsh, just east of the Timber Point Marina which abuts the Timber Point Golf Course in Great River and Johns Neck marsh on the Unchachogue Creek in Mastic.
- Havens Point, where no pesticide spray takes place, was used as the control or reference site.

Johns Neck and Timber Point marshes were assessed in the beginning of the week for the presence of mosquito larvae. The method for larvae collection calls for the use of a mosquito larvae dipper. With the dipper, mosquito larvae are sampled using the standard “dip count” method, taking care not to cast a shadow onto the standing water that is being sampled; otherwise, the mosquito larvae will quickly disperse. The edges of standing water were sampled in order to get a full dipper of water. Each sample was saved in mason jars with a mosquito

larvae field report placed around the sample. The samples are brought back to the ABDL for identification purposes and to verify the count of the number of larvae per sample.

Altosid® and Vectobac 12 AS® (*Bti*) were used as a duplex mix in two operational aerial sprays on August 3, 2004 and August 10, 2004. Altosid® was used alone on a third operational spray on September 1, 2004. The delivery amount of the active ingredient of Altosid® or s-methoprene is one oz./acre and the active ingredient of Vectobac 12 AS® or *Bti* is delivered in the amount of one pint/acre.

Two to three days post spray, mosquito larvae were collected and stored in mason jars. Every attempt was made to bring the larvae back to the laboratory alive, in order to track emergence rates. The larvae were counted and identified by species. The larvae were placed in larvae containers and monitored daily (Figure 2).

3.3 Results

August 3 Larvicide Spray at Johns Neck and Timber Point using Havens Point as reference site: Altosid used in conjunction with *Bti*.

Note: staff applied VectoLex CG® by ground application in the Timber Point golf course area on the same day as the helicopter spray.

Table 2 lists the emergence results from the three aerial larvicide sprays. In summary, for the first spray, no adults emerged from the Timber Point or Havens Point sites. Mosquito larvae were collected from all three sites and all were identified as stage III or stage IV *Ochlerotatus sollicitans* larvae. From the Timber Point site, one stage III larvae did pupate, but was found dead.

At Johns Neck, from 12 larvae (seven Stage III, five Stage IV) collected, one pupa appeared (8.33 percent pupation rate) for the total of four samples collected. From one of the Johns Neck sample, one adult did emerge which could have originated from the one pupae collected in that sample or from the two larvae, one of which had changed into the pupal stage. At Havens Point, one pupa did develop from the six larvae (a pupation rate of 16.67 percent) from the total of four samples collected there.

August 10 Larvicide Spray at Johns Neck and Timber Point with Havens Point as reference site- Altosid used in conjunction with *Bti*

The caged aquatic organism site at Timber Point was moved, which did not change where the mosquito larvae samples were discovered. Once again, no mosquito larvae were found close to the caged aquatic organism sites. The Timber Point marsh had been recently flooded, with many fish observed high on the marsh and no larvae and one pupa collected. The only mosquito sample collected from all three sites was one live pupa from Timber Point, which did not further emerge into the adult stage.

September 1 Larvicide Spray at Timber Point with Havens Point and Johns Neck used as reference sites-Altosid used alone

Larviciding did not occur at the Johns Neck caged organism site but did so due east of this location. Large numbers of larvae were collected at both Timber Point and Johns Neck, but not in the immediate vicinity of the caged aquatic organisms. The larger numbers of larvae collected for this trial was most likely due to the fact that no *Bti* was used, so that the larvae that were targeted did not die from the treatment before being collected.

The majority of the larvae collected were *Ochlerotatus sollicitans*, with a few *Oc. taeniorhynchus* collected at both Timber Point and Johns Neck and one *Oc. cantator* larvae collected at Havens Point on September 1, 2004. The four Timber Point samples resulted in a total of 174 stages II-IV larvae collected. No pupae were collected. 16 healthy pupae developed and 4 healthy adults emerged. The rate of emergence is 2.3 percent or four adults/174 larvae. This translates into a 9.2 percent pupation rate or 16 pupae/174 larvae. The rate of pupal development into adults is 25.0 percent or four adults/16 pupae.

From the Johns Neck samples, approximately 104 larvae from all four larval stages were collected from a site due east of the caged organisms in two samples. No pupae were collected here. The pupation rate for larvae into pupae is 2.9 percent or three pupae/104 larvae. No further development of the pupae occurred.

From Havens Point, the one larvae found was identified as a Stage III *Oc. cantator* which did eventually develop into an adult mosquito for an emergence rate of 100 percent (albeit from a very small sample size).

3.4 Discussion

It appears that there is a good reason why Havens Point is not sprayed, since there were so few mosquito larvae collected on three separate days which led to very few larvae being available as controls. In areas adjacent to the caged organism sites, which were also part of the spray swath, larvae and pupae were collected from Timber Point and Johns Neck. When only Altosid® was used on September 1, 2004, the numbers of mosquito larvae collected was much higher than the two other sprays. *Bti* used on the other spray dates may have killed many larvae. The failure of most larvae to pupate after Altosid® exposure could be due to the material or to other factors, such as availability of suitable food or other conditions while they were held in the laboratory. When larvae were able to pupate, few adults emerged successfully. This would support the idea that the treatments were successful in delivering effective methoprene doses to the cage sites.



Figure 2 - Lab container for mosquito larvae with mosquito collection jar in bottom half of container

Table 2 – Mosquito Larvae

Date of Spray: 8/3/04	TP	TP	TP	TP	TP	TP	TP	TP	TP	TP	
Larvae # and Condition	0	10	0	0	3 Alive	1 Alive	1 Alive	9 Dead	5 Alive	2 Dead	26 Dead
Larvae Stage	--	Stage IV	--	--	Stage IV	Stage III	Stage III	Stage IV	Stage IV	Stage III	Stage IV
Pupae developed	0	0	0	0	0	1,D	0	0	0	0	0
Adults emergent	0	0	0	0	0	0	0	0	0	0	0
Percent (%) adult emergence	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%
Percent (%) pupation	0%	0%	0%	0%	0%	100.0%	0%	0%	0%	0%	0%
Date of Spray: 8/3/04	JN	JN		JN	JN	JN	HP	HP		HP	HP
Larvae # and Condition	0	5 Alive	5 Alive	1 Dead	1 Dead	2 Alive	0	1 Alive	1 Alive	2 Alive	2 Alive
Larvae Stage	--	Stage III	Stage IV	Stage III	Stage III	Stage III	--	Stage III	Stage IV	Stage II	Stage III
Pupae developed	0	0	0	0	0	1	0	0	0	1	0
Adults emergent	0	0	0	0	0	0	0	0	0	0	0
Percent (%) adult emergence	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%
Percent (%) pupation	0%	0%	0%	0%	0%	50.0%	0%	0%	0%	50.0%	0%
Date of Spray: 8/10/04	TP	JN	HP								
Larvae	0	0	0								
Pupae developed	0	0	0								
Adults emergent	0	0	0								
Percent (%) adult emergence	0%	0%	0%								
Percent (%) pupation	0%	0%	0%								
Date of Spray: 9/1/04	TP* (1 of 2)	TP* (2 of 2)	TP* (1 of 2)	TP* (2 of 2)	TP	TP	JN	JN* (1 of 2)	JN* (2 of 2)	HP	
Larvae # and Condition	100+ Alive		50+ Alive		10 Alive	14 Alive	4 Alive	100+ Alive		1 A live	
Larvae Stage	<u>Stages II, III, and IV</u>		<u>Stages II, III, and IV</u>		<u>Stage III</u>	<u>Stages II, III, and IV</u>	<u>Stages 1 and II</u>	<u>Stages II, III, and IV</u>		<u>Stage III</u>	
Pupae developed	0	0	6 (2 dead)	7	2	3	0	1	2	1	
Adults emergent	0	0	4 (2 dead)	2 (1 dead)	1	3 (3 dead)	0	0	0	1	
Percent (%) adult emergence	0%	0%	6.0%		10.0%	0%	0%	0%	0%	100.0%	
Percent (%) pupation	0%	0%	26.0%		20.0%	21.4%	0%	3.0%		100.0%	

Key: TP - Timber Point in Great River, Spray site

JN - Johns Neck in Mastic, Spray site

JN - Johns Neck in Mastic, control site that was not sprayed ONLY on 9/1/04

HP - Havens Point in East Moriches, control site that was not sprayed

* The samples were split using only the water the larvae were collected in.