6 Early Action Projects

One of the reasons for conducting the Management Plan process as part of this process was to obtain local information to inform the environmental assessment process. Although the generation of new information is normally not part of an environmental review under SEQRA, in this instance, many projects were undertaken to generate information regarding water management and pesticide impacts in Suffolk County. Approximately $900,000 of the $3.5 million consultant budget (more than 25 percent) was reserved for these kinds of projects. This excludes the characterizations of the Risk Assessment Areas (Section 4) and the Primary Study Areas (Section 5) discussed earlier.

This section will present the results of the major efforts that have been completed or are well underway at this time:

- Wertheim National Wildlife Refuge Open Marsh Water Management (OMWM) Demonstration Project (Wertheim)
- Impacts to Caged Organisms from Vector Control Pesticides Experiment (Caged Fish)
- Wertheim National Wildlife Refuge-Seatuck National Wildlife Refuge Marsh History Determination Project (Wertheim-Seatuck Retrospective)
- Mosquito Ditch Conveyance of Pollutants Experiment (Runoff Experiment)
- Keystone Marsh Invertebrate Species and Larvicides Survey (Non-Target Invertebrates)
- Spotted Turtle Research in Napeague Marsh (Turtles)
- Catchbasins Mosquito Breeding Survey (Catchbasins)
- Recharge Basin Breeding Survey (Recharge Basins)
- Non-standard Control Measures Efficacy Tests (Alternatives)

In addition to these efforts, the Long-Term Plan also intends to conduct a test of the ability to use satellite imagery to track types of vegetation in salt marshes; and demonstrate the potential to track marsh vegetation trends over time using this technology. This Remote Sensing project was delayed by contractual difficulties, and has just been initiated early in 2006. The County is hopeful that its primary researcher for this project, Kamazima Lwiza, PhD of the Marine Sciences Research Center, Stony Brook University, acting as a subconsultant to CA (and with CA assistance), will be able to complete this project before the end of 2006.

6.1. Wertheim National Wildlife Refuge Open Marsh Water Management Demonstration Project

As discussed in Section 5, Wertheim National Wildlife Refuge, located at the mouth of the Carmans River (on the south shore of Suffolk County) (Figure 6-1), has been the site of an ongoing demonstration project for OMWM. USFWS is the project sponsor and permit holder for this project. CA has been project manager, but the execution of the project has required close collaboration with USFWS, Ducks Unlimited (as a subconsultant to CA), SCVC, and SCDHS. In addition, technical input and assistance had been received from Steven Goodbred, PhD, Marine Sciences Research Center, Stony Brook University, and his laboratory (as a subconsultant to CA) (Dr. Goodbred is now at Vanderbilt University, and so is not as readily available), Paul Capotosto, CTDEP, Susan Adamowicz, PhD, USFWS Region 5, and Steven Terracciano of USGS, Long Island Subsection (acting as a collaborator with SCDHS). NYSDEC, especially the Natural Resources Division and the Permits Division, also worked closely with project designers.

USFWS is developing a policy that will intend that pesticide use be eliminated wherever and whenever possible on its refuges. USFWS also has a policy that its refuges should be "good neighbors," whenever possible. The Wertheim NWR can generate mosquito broods that then infest surrounding residential areas. Wertheim managers are interested in determining if alternatives to larvicides can be employed to limit mosquito breeding at the site to comply with both policies. Additionally, Wertheim managers have a mandate for the refuge to maximize water fowl habitat and, therefore, water fowl use of the site.

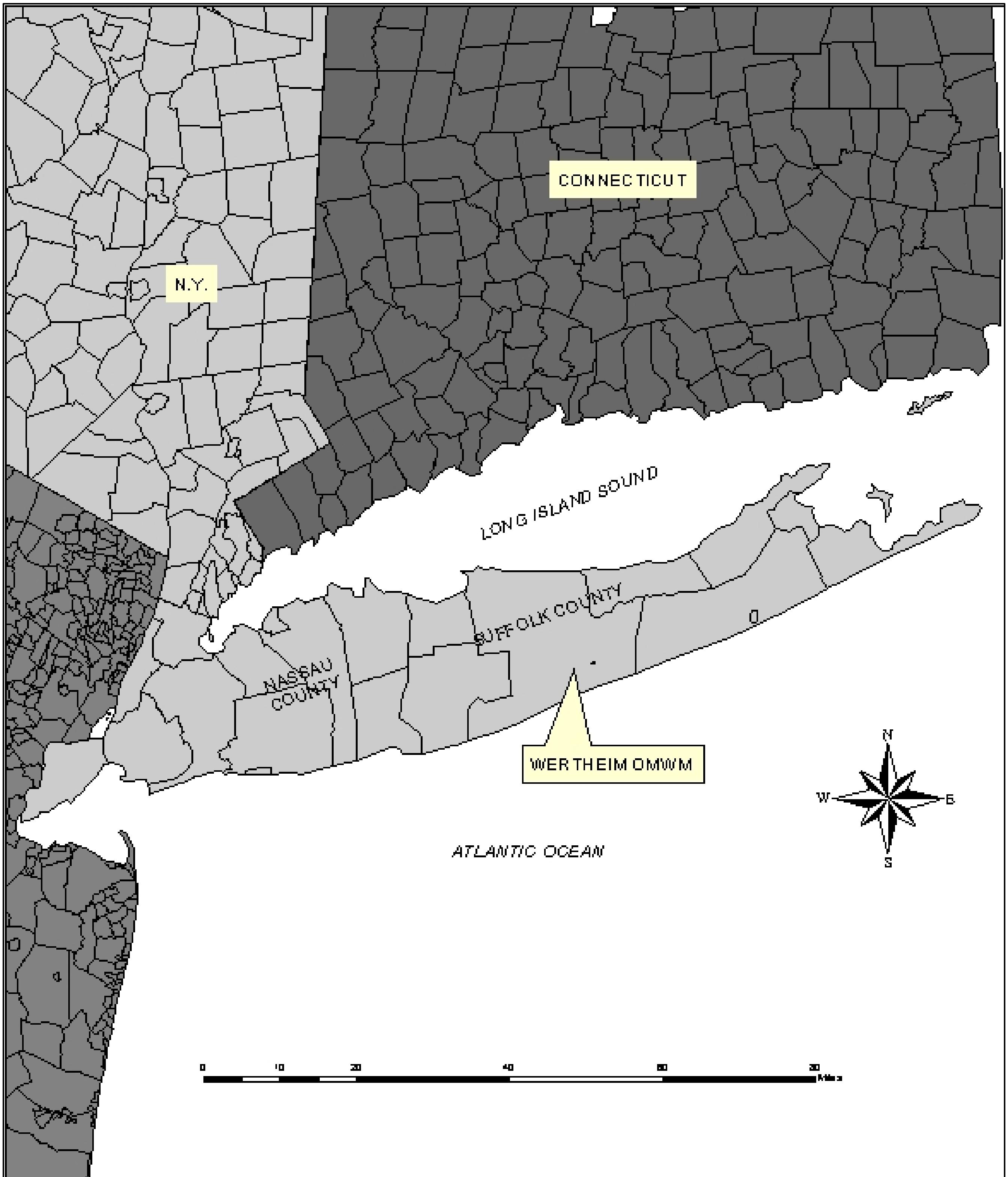


FIGURE 6-1
DRAFT GENERIC ENVIRONMENTAL
IMPACT STATEMENT
WERTHEIM NATIONAL WILDLIFE REFUGE
SITE LOCATION

Mosquito control literature generally finds that grid-ditching reduces water fowl use of a marsh (Bourn and Cottam, 1950; Clarke et al., 1984), although a few studies have disputed the point (Corkran, 1938; Provost, 1977). Wertheim NWR managers allowed ditch plugs to be installed across many of the mosquito ditches in the late 1980s and throughout the 1990s, to address potential ditching impacts and still maintain mosquito control. Many of these plugs failed, mostly due to muskrat undermining, and comprehensive evaluation of the impacts of OMWM efforts was never made. In 2001, USFWS and USGS jointly began research that was intended to determine the impacts of ditch plugging on marshes throughout USFWS Region 5 (from Maine to Delaware). The study design called for a BACI (Before-After, Control-Impact) approach to be followed. However, no new plugs were installed as part of the project, and, in fact, at all of the sites across Region 5, the plugs had been installed prior to the study initiation in 2001. Control (unplugged) sites were found for each site (James-Pirri et al., 2001). Wertheim NWR then became a study site. Although the use of control sites could help determine if impacts occurred, or benefits accrued, the study's power appeared compromised from the start because of its inability to precisely implement the BACI design.

Ducks Unlimited arranged for USFWS to meet with SCVC regarding using Wertheim as a demonstration project site. A preliminary design was created for the site, and was included in early workplan drafts. The original concept was to test variations of ditch plugs at several large areas along the east bank of Wertheim. Four sites would be needed to test

- full ditch plugs
- "sill" ditch plugs (plugs that allow for some tidal exchange because they do not reach the marsh surface elevation)
- runnels (an Australian technique involving construction of shallow ditches that are not intended to alter site hydrology)
- one area to serve as a control site

Therefore, four experimental areas (Areas 1 through 4) were identified for the project (Figure 6-2).

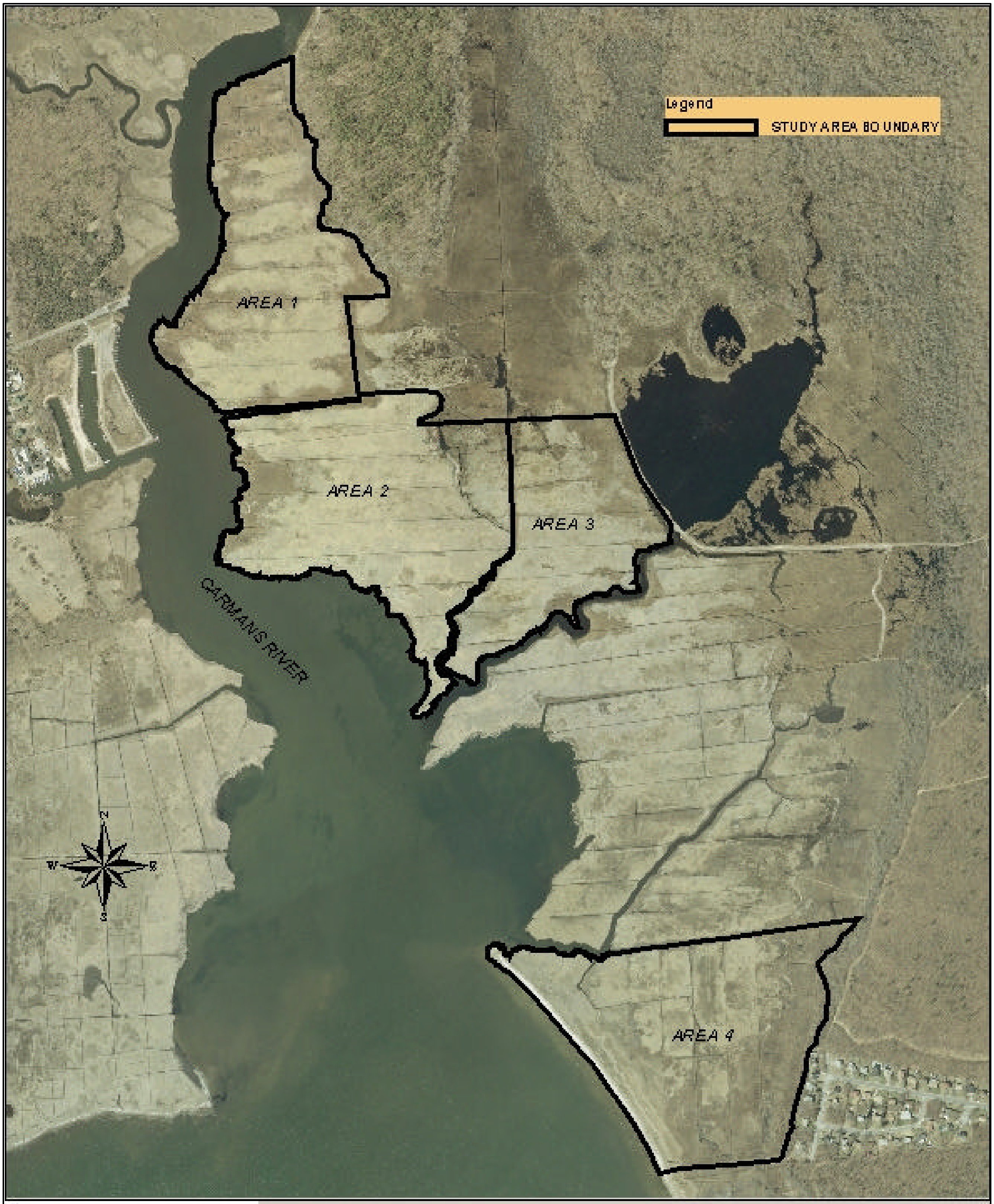


FIGURE 6-2
DRAFT GENERIC ENVIRONMENTAL
IMPACT STATEMENT
WERTHEIM NATIONAL WILDLIFE REFUGE
AREAS 1,2,3 & 4

This method would clearly allow for a BACI approach to the effects of the manipulations. The USFWS-USGS study site occupies the area between Area 3 and Area 4 along the east bank of the river.

Permit negotiations were entered into with NYSDEC. NYSDEC had great concerns regarding monitoring, which occupied much of the initial discussions. These concerns had two foci:

- One focus was on the extent and commitment to monitoring. Previous OMWM demonstration projects had not collected appropriate information, with the exception of the Seatuck demonstration project (Lent et al., 1990). NYSDEC therefore had no means to assess claims made regarding OMWM successes or failures. Even the Seatuck project had minimal follow-up measurements of the impacts of the work. NYSDEC therefore pressed for a complete monitoring approach to determine if any impacts would result from the project. Typical areas of interest would be the use of the marsh by fish and birds, changes in invertebrate populations, and effects on water quality and hydrology, and vegetation productivity and speciation.

- NYSDEC was also concerned that the variability of weather and marsh processes could make any monitoring data contingent on temporal variations rather than project impacts. Therefore, NYSDEC expressed an interest in at least five years of pre-project monitoring, with similar post-project efforts.

Project participants were hesitant to accept this onerous monitoring regime which would delay the start of the early action demonstration project. Funding available through the Long-Term Plan was intended for a three year project, and did not include monitoring to the extent called for by NYSDEC. Additionally, USFWS and USGS recently completed a three year monitoring effort at Wertheim, also intended to determine impacts of OMWM on ditched marshes (James-Pirri et al., 2002). Because of the similarities in monitoring techniques and the geographical closeness of the two efforts NYSDEC allowed USFWS/USGS data to be considered in this project.

In the fall of 2003, CA (with Ducks Unlimited, assisted by the County and USFWS) began a comprehensive monitoring program across Areas 1 through 4. Transects were identified across each Area, using the USFWS/USGS protocols (James-Pirri et al., 2002). Twenty-four stations were established in each of Areas 1 and 2, and 20 stations were established in Areas 3 and 4, for a total of 88 marsh surface stations. In addition, 10 ditch sampling points (fish stations) were established in each Area (a total of 40), again using the USFWS/USGS protocols. SCDHS also established four permanent Carmans River water quality monitoring stations (one associated with each Area), and two water quality monitoring stations in navigable sections of the major creeks. Tables 6-1 through 6-3 list the monitoring program adopted for the project. In addition to the listed efforts, vegetation across the four areas was characterized in 2003 by field-truthing aerial photography interpretations. Mosquito breeding locations across Areas 1 and 2 were completely characterized in the summer of 2004, and the extent of inundation across Areas 1 and 2 was determined for a spring high tide (July 2004).

Table 6-1. Biological Parameters

Parameter	**Responsible Party**	**Assisting Parties**	**Locations**	**Frequency**	**Technique**
Mosquito Breeding Concentration Areas	DU	SCVC	All four areas	Until 9/15	Traverse marsh, visually identify breeding location, flag
Mosquito Dip transects	DPW	DU	All transects stations (88)	Weekly through October	Take dip samples at locations near/at stations
Mosquito breeding	SCVC	DU, USFWS	Areas 1 and 2	Summer 2004	GPS mosquito breeding sites
Vegetation quadrats	DU	USFWS	All transect stations (88)	Once by end of October	USFWS/USGS manual (speciation primarily)
Fish sampling	DU	SCVC & SCDHS CA USFWS	All fish stations (40)	Three times a year, May, July, Septemberr	USFWS/USGS (nets)
Invertebrates	DU & CA		Surface: 26 samples (stratified by cover) Water column/benthos – 70% of fish stations	Once a year	At transects: USGS surface core; at fish, 1 meter net twirl
Invertebrate sample analysis	CA				Initial processing, Abundance, biomass, identification (to family)
Vegetation biomass	CA		Surface clip (50% -- 44 stns) Clip & core (25% -- 22 stns)	Once a year, by mid-October	Root & stalk within dm, dried mass
Marsh composition	DU	USFWS DU surveyor	All four areas	end of the year	Groundtruthed aerials; surveyor
Birds (initial)	DU				Anecdotal observations
Birds (later)	DU		All four areas	3x/yr	Formal survey techniques

Table 6-2. Physical Parameters

Parameter	**Responsible Party**	**Assisting Parties**	**Locations**	**Frequency**	**Technique**
Ditch qualities	DU	USFWS	All 4 areas, all ditches	once	Visual inspection, measurement (plug status, deep areas, avg. widths, terminus, intersections, etc)
Water flows	DU	CA	All ditches	Several (varying tidal conditions)	Drift cards
Fresh water sources	DU		All ditches (upper ends)	Low tides	Visual inspection for "dry" ditches; salinity meter for fuller ditches
Sedimentation	CA		All transect stations (88)	Once a year	Marker horizon
Water table height	DU	DU surveyor	All transect stations (88)	every 10-14 days	Measure from surveyed top-of-casing (decision needed about this)
Water table height – relative fresh water inputs	DU	SCDHS (installation)	Upland monitoring wells		Measure from surveyed top-of-casing (electronic meter)
Visual changes	CA		Across each area , to cover each completely (~40 in total)	Immediately post-construction, then once in September	Elevated (ladder) photo stations
Marsh Inundation	CA	DU, SCVC, SCDHS, USFWS	Areas 1 & 2	Once (July 2004)	Glue sticks

Table 6-3. Chemical Parameters

Parameter	**Responsible Party**	**Assisting Parties**	**Locations**	**Frequency**	**Technique**
Carmans River WQ	SCDHS		4 stations	3x/year (approx. quarterly	Std.; full SCDHS parameter list
Salinity, T, Cond., pH, DO	SCDHS	DU	Deep portions of ditches	Rotate on bi-weekly basis	YSI continuous sondes (may deploy several)
Ditch salinity surveys	DU	SCDHS	All ditches	At least once, more is better	Every 50 m or so, measure salinity in ditch
Water table/pore water salinity	DU		All transect stations (88)	every 10-14 days	YSI meter
WQ parameters (Sal, T, Cond, pH, DO)	SCDHS	DU	All fish stations (40)	~ Bi-weekly but rotate through tidal cycles	YSI meter plus pH meter
Nutrient sampling	SCDHS		Selected fish stations (12 total)	2x/yr (June/August)	Random selection from fish stations; SCDHS nutrient sampling protocols

The Suffolk County Legislature appropriated supplemental funding to support this monitoring effort and other consultant work associated with the site in August 2004.

Negotiations regarding the length of pre-project monitoring followed the initiation of monitoring. One means of addressing NYSDEC concerns was a project commitment to use all relevant USFWS/USGS data generated in the immediate vicinity of the project site.

As monitoring continued into 2004, research continued on the different means of conducting OMWM. These efforts, along with literature searches, included field trips to New Jersey project sites and presentations by Paul Capotosto on Connecticut techniques and means of developing projects.

A grand design collaboration, lasting two days, was conducted in May 2004. One day was spent largely in the field, and the other was spent at SCDPW offices. Susan Adamowicz and other local USFWS personnel, Craig Kessler and other Ducks Unlimited personnel, Steven Goodbred and members of his laboratory, Paul Capotosto, CA, SCDHS, and SCVC all met to determine the optimal design for the site. This process determined that ditch plugs were unlikely to provide the degree of mosquito control and the natural resource enhancements needed by USFWS. An alternate approach to the design focused on enhancing water circulation through the marsh as a means of generally improving water quality to support killifish, and also to potentially control invasive *Phragmites.* Installing ponds in areas where mosquito breeding was most intense was identified as a preferred option. The ponds would be large enough to serve as attractive water fowl habitat. Spoils from pond construction would not only be used to fill tussocky mosquito breeding areas in the high marsh, but also to fill most of the existing grid ditches. The project had become a major marsh restoration as well as a mosquito control demonstration project.

NYSDEC was pleased with the overall approach to the project as marsh restoration, instead of just an alternative form of mosquito control. Designs were finalized over the course of the summer, and a permit application prepared in fall 2004. NYSDEC reviewed the application and issued a permit ahead of schedule in January 2005, for construction to begin immediately.

Unfortunately, paperwork with the USACOE was not processed as expeditiously. USACOE determined that the general national permit sufficed for the project, but not until the end of

February 2005. As construction was to cease April 1, because of the onset of bird nesting at Wertheim, this allowed only one month for construction activities. For this reason, only the design for Area 1 was implemented then. Area 2 was constructed instead from late January through March, 2006 (Areas 3 and 4 serve as control sites). Construction in Area 1 was completed on March 30, 2005 (Figure 6-3 shows the implemented design) (an as-built drawing for Area 2 had not been prepared as of April 15, 2006; Figure 6-4 shows the design lay-out for Area 2).

Anecdotal evidence shows that the project has been a major success. The marsh revegetated over the summer, 2005. Mosquito breeding was nonexistent until the end of the summer, where certain parts of the project supported larvae (predominantly in areas on the edge of the project and in some of the filled ditches). These areas were touched up in March 2006, as part of the Area 2 construction activities (the short time available for construction in March 2005 meant that not all aspects of the construction were as polished as might have been desired). Water circulation has been brisk (visually). Waterfowl use of the ponds, in spring and early summer, was notable, and shorebirds foraged extensively through Area 1 all summer. Blue crabs were found in the constructed waterways of the marsh; the ponds were well-stocked with killifish, and *Menidia* and sheepshead minnows have also been well represented in the ponds. A clapper rail was found nesting in Area 1 in July, and a sea robin was spotted in the main tidal channel. Beneficially, *Phragmites* regrowth appears to be stunted in several areas.

Monitoring has continued, and should document these successes. Fish stations in Area 1 needed to be relocated with the loss of ditches, and a new fishing technique is needed for the ponds (the USGS-USFWS protocols do not work with an uneven pond bottom).

More time is needed before a final determination of project success can be made. It was a very wet spring and an extremely dry summer in 2005, which may have had impacts on the conditions at the site. Some of the natural resource use of Area 1 may have to do with the transition from unvegetated to vegetated marsh following the travails of construction. Nonetheless, the first impression of the project is that it will meet the expectations of USFWS for natural resource augmentation and mosquito control.

At this time, no analysis of the collected data has been made. Expectations are that, at the end of the 2006 monitoring season, CA will provide preliminary analyses of the four years of monitoring data available for Areas 1 through 4, and determine if statistically significant changes occurred across time and/or space at the site. Per the NYSDEC permit, annual monitoring reports have been prepared (Cashin Associates, 2006a, is a three year compilation).

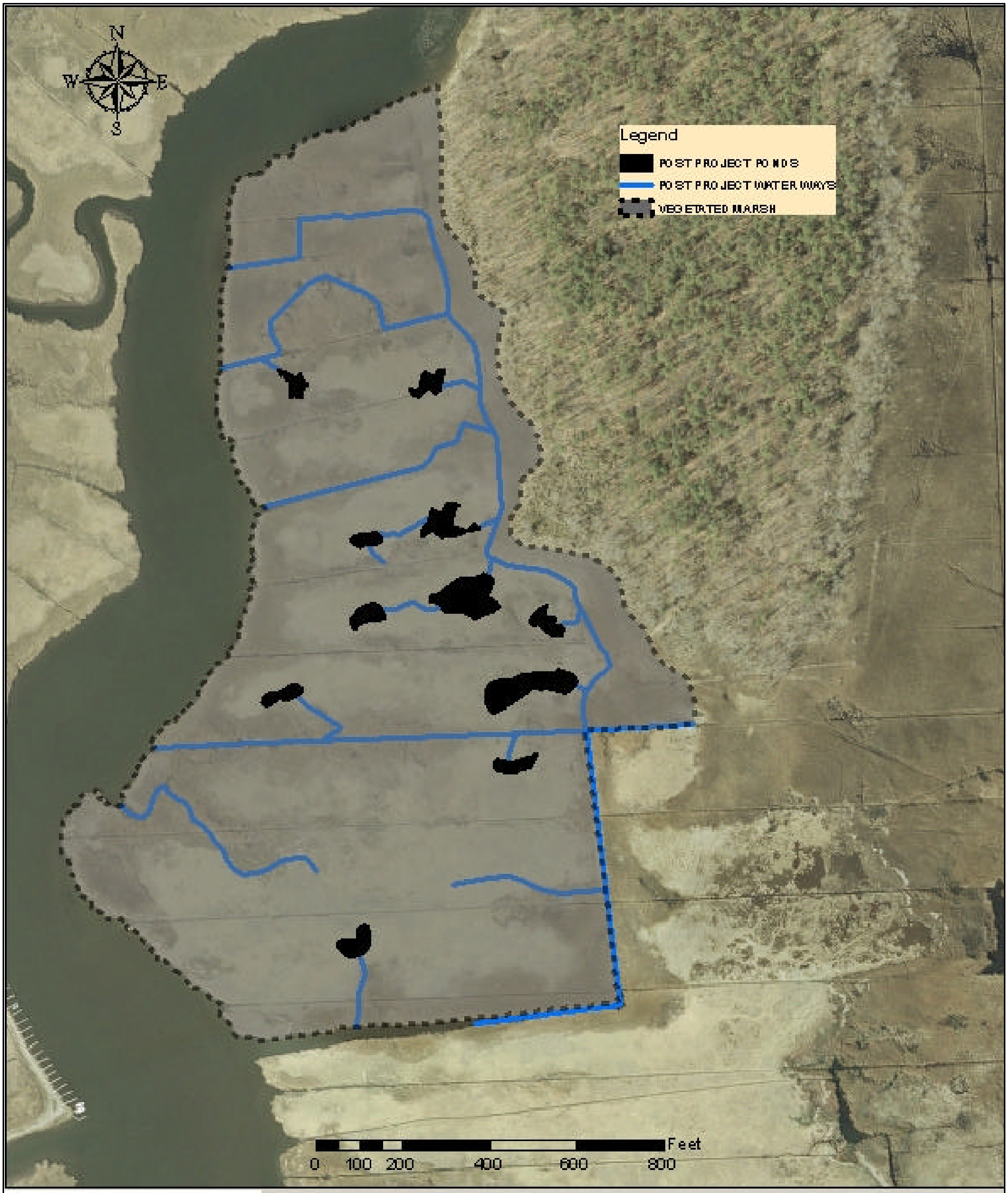
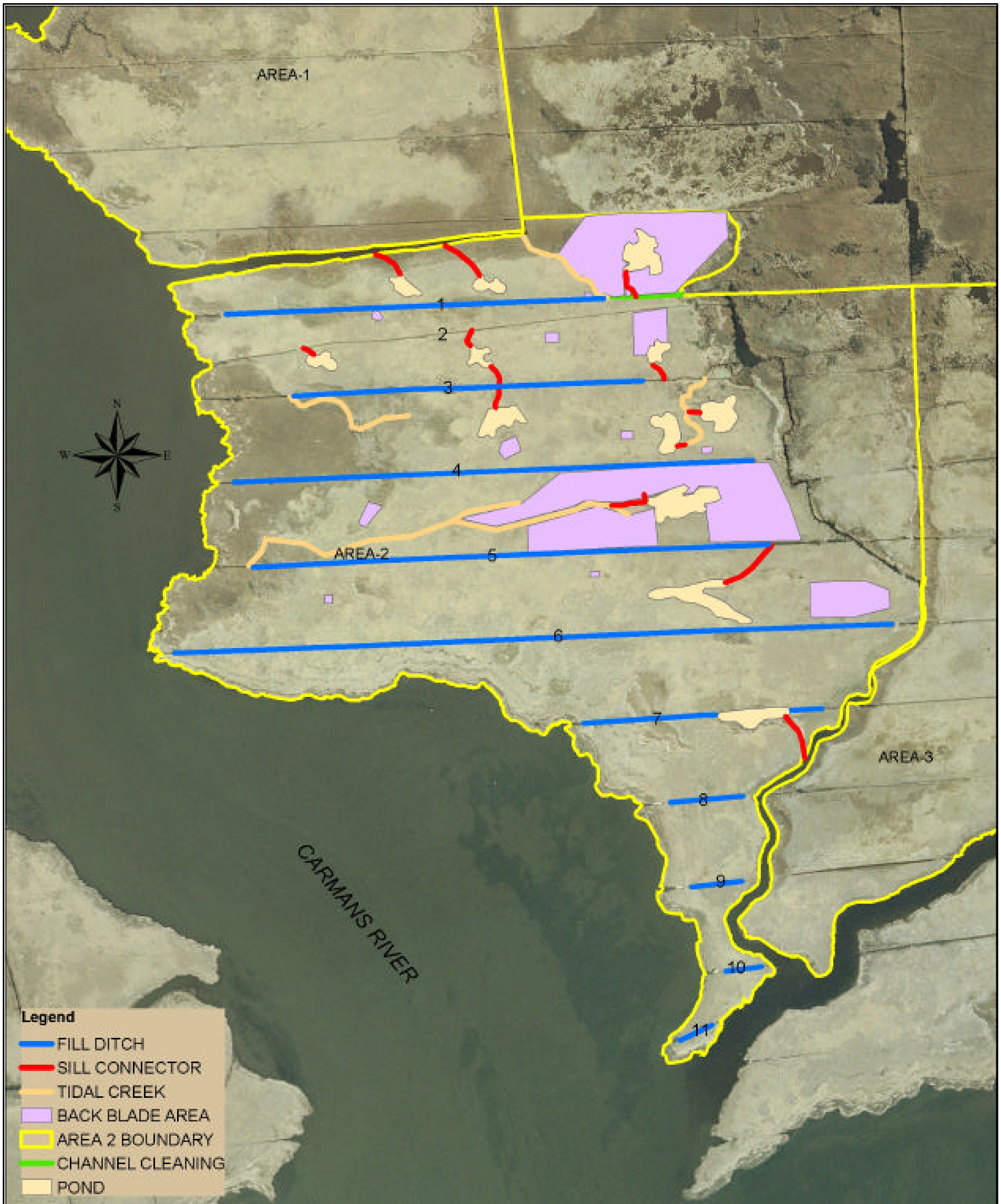


FIGURE 6-3
DRAFT GENERIC ENVIRONMENTAL
IMPACT STATEMENT
WERTHEIM NATIONAL WILDLIFE REFUGE
AREA 1 POST PROJECT



Cashin Associates, P.C.
ENGINEERING · PLANNING · CONSTRUCTION MANAGEMENT

SUFFOLK COUNTY VECTOR CONTROL
AND
WETLAND MANAGEMENT LONG-TERM PLAN
DRAFT GENERIC ENVIRONMENTAL IMPACT STATEMENT
FIGURE 6-4
AREA 2 CONSTRUCTION PLANS

6.2. Impacts to Caged Organisms from Vector Control Pesticides Experiment, Part 1: Impacts to Biota

6.2.1. Background Information

One comment received in the DGEIS Scoping was from NYSDEC, and requested that the project undertake a particular experiment that has become known as the Caged Fish experiment. The NYSDEC suggested protocol was for tests of at least two local organisms in both fresh and salt water, where the organisms would be subjected to applications of adulticides. The organisms would be observed for at least 24 hours to determine immediate and delayed mortality. Sets of organisms would also be placed at control sites (where no chemicals had been applied). NYSDEC also supplied a statistical methodology to be used to determine the significance of the results.

The Long-Term Plan produced a Responsiveness Document as part of its Scoping effort. In it, the Caged Fish experiment was addressed as follows:

> Despite substantial and substantive criticisms of the caged fish experiment, the County will work with NYSDEC to create a jointly-acceptable field test of acute toxicities associated with a selection of adulticides (and, potentially, certain larvicides).

(CA-CE, 2002)

The criticisms alluded to in the Responsiveness Document included comments offered by Edward Nadel, a biostatistician in SCDHS, which were included in Scoping, and solicited comments from Anne McElroy, PhD, Stony Brook University (serving as a subconsultant to CA).

For most of 2003, due to certain funding problems, the Long-Term Plan process was not conducted at full-strength. Some activities relating to the Caged Fish experiment did occur, however.

Cornell Co-operative Extension was requested by CA to develop a Caged Fish experiment, based on the requested NYSDEC protocol but also addressing comments made by McElroy and Nadel. Cornell Co-operative Extension, recognizing that certain methodological problems were required to be addressed prior to the completion of such a workplan, produced a generalized approach and a budget based on the maximum number of likely experimental sites and iterations. CA used this as a basis for a supplemental funding request to the Long-Term Plan managers, in order to address changes in the project due to Scoping.

The Suffolk County Legislature, who approved the overall project funding in April, declined to directly fund the experiment. This was because the Legislature hoped that SCDHS would be able to secure funding from either Federal or State sources. Therefore, in September, SCDHS prepared (with the assistance of CA) a grant proposal to the New York State Environmental Quality Bond Act (EQBA) fund. The proposal was still based on the rather rough workplan prepared by Cornell Co-operative Extension, although Dr. McElroy and CA had succeeded in addressing some of the more problematic issues. The proposal, based on Cornell Co-operative Extension estimates, called for leveraging other Long-Term Plan activities with the proposed EQBA funds to cover the costs associated with the experiment.

A Long-Term Plan “Monitoring” subcommittee was formed, and had several meetings in 2003. Members of this committee included CA, SCDHS, SCVC, USGS (Steve Terracciano), NYSDEC, and various members of the CA subconsultant team including Ken Skipka (RTP Environmental), Bruce Brownawell, PhD (Stony Brook University), and members of Cameron Engineering. Discussions that related to the project included:

- The dry deposition experiments conducted by the SCDHS Office of Ecology under the direction of Ken Hill (PEHL)
- Means of relating various data sets collected by Dr. Brownawell, USGS, and PEHL
- Use of models (including one developed by Adapco) to determine the area impacted by aerial applications of insecticides

Following Dr. McElroy’s initial reworking of the Cornell Co-operative Extension proposal as part of the EQBA application process, it became clear to CA and SCDHS that the project had become more complex than originally envisioned. Dr. McElroy was asked by CA to assume the lead technical role on the project.

During the summer of 2003, unknown to the Long-Term Plan, students under the direction of Southampton College faculty conducted a Caged Fish Exposure to Insecticides experiment. The organisms were exposed to larvicides and adulticides. The data was assessed in the fall, and a report was released in the spring of 2004 (SCERP, 2004).

In early 2004, the Long-Term Plan learned that the State would decline to fund the experiment. The funding applications were submitted to the Suffolk County Quarter Percent Committee, which recommended funding the projects. In April, the Legislature approved the funding proposals. There were some problems with the form of the approval that impacted the delivery of funds to the project, but there were no material impacts from these problems on the experiment itself.

Dr. McElroy drafted another copy of experimental procedures for the Caged Fish experiment due to the preparation of materials for the Legislature. These procedures were circulated among interested parties (SCDHS, SCVC, Cornell Co-operative Extension, USGS, other CA subconsultants, and NYSDEC).

On March 19, 2004 another meeting of the Monitoring Committee was held. A frank discussion of the Caged Fish experiment procedures occurred between CA and NYSDEC. At this time, CA proposed the use of controlled sprays as a means of dosing the organisms. NYSDEC expressed a desire to see a proposal that used operationally-required applications as the basis for the proposal. NYSDEC also preferred to see artificial fresh water environments (golf course ponds or recharge basins) used for that portion of the experiment, to avoid impacts to natural water bodies. NYSDEC pointed out that Article 24 appeared to allow for the possibility of waiving the requirement for operational applications, if a case could be made that a waiver should be granted for a demonstration or experimental purpose.

The project proposal was therefore reconsidered. Environments such as Timber Point golf course, where salt marshes and freshwater sites are found in close proximity, were very carefully considered. However, Dr. McElroy also counseled that the need for control sites and appropriate replication made it unlikely that two different tests could be successfully managed at once.

At this time, the Southampton College report was released (SCERP, 2004). Robert Turner and Chris Gobler of Southampton College were invited to join the project team. Perceived problems with the Southampton College work made the need for adequate replication and supporting data collection even more important. The project scope was expanded so that it now included tests of methoprene and resmethrin, extensive water and sediment sample collection (Bruce Brownawell and USGS), caged mosquito and larval impacts (SCVC), air sampling (PEHL), meteorological data collection to support air modeling (RTP, potentially with Adapco, a private modeling company), and dry deposition monitoring (SCDHS Office of Ecology). Two sites would be used, both for control samples as well as for exposures. The organisms (sheepshead minnows and grass shrimp) would be evaluated for mortality. The minnows would be tested for growth rates and the shrimp would be evaluated for prey capture and fecundity. The experiments would be repeated twice, for both chemicals.

This proposal was submitted for NYSDEC consideration. On June 18, 2004, a meeting was held at NYSDEC Region I to discuss the proposal. NYSDEC accepted most of the proposal, but held fast to its need for the insecticide applications to occur on an operational basis. This would not be a problem for methoprene, as SCVC applies this larvicide on nearly a bi-weekly schedule to many salt marshes across the County; however, SCVC had only made aerial applications of resmethrin six times over the preceding five years, all for heath emergency reasons. Further discussion resulted in provisional NYSDEC acceptance of the alteration of truck-based adulticide applications, which are how vector control is achieved, to aerial applications (NYSDEC has generally not allowed aerial applications, which usually require release from Freshwater Wetlands Regulations which restrict applications of pesticides to fresh water wetlands, to be conducted for vector control reasons). The acceptance was provisional based on a final interpretation of the Freshwater Wetlands Regulations. CA was to submit an application for waiver of the regulations on the "experimental" clause in Article 24 (the aerial applications necessitated a fresh water permit decision). On July 2, 2004 CA submitted the waiver request.

NYSDEC was unable to waive the fresh water regulations. However, in response to a request by CA, it did issue a permit for the aerial adulticide applications on July 15, 2004. The permit allowed SCVC to apply resmethrin via helicopter at Gilgo Beach or Mastic-Shirley, in place of operationally-required truck-based applications.

The discussion of regulatory issues and project evolution is more completely presented in Cashin Associates (2005a).

The experiments were conducted through August (see below). Three larvicide and two adulticide applications were tested. Some related work, such as sampling benthic invertebrates to determine if long-term impacts from larvicides could be discerned, and laboratory work in conjunction with the field work, was conducted in the fall of 2004. All work on the project has been completed.

The work was funded by the County Legislature. CA was project manager. Cameron Engineering, RTP Environmental, and Stony Brook University (both through the State University Research Foundation and through direct arrangements with individual research laboratories) were subcontracted to CA. Southampton College indirectly subcontracted with CA, through the State University Research Foundation. USGS had a cooperative agreement with SCDHS.

Project Participants

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Simonette Rivera

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Barbara Cardenas
Brian Aerne

6.2.2. Tests for Impacts to Biota

This section of the Caged Fish experiment, is drawn from Cashin Associates (2005b), unless directly cited differently.

Methods:

Cage Deployment:

Aerial application sites used in the experiment were Timber Point and Johns Neck, with Havens Point and Flax Pond as reference sites (Figure 6-5). The study was partially confounded by low oxygen and high temperature during test deployments.

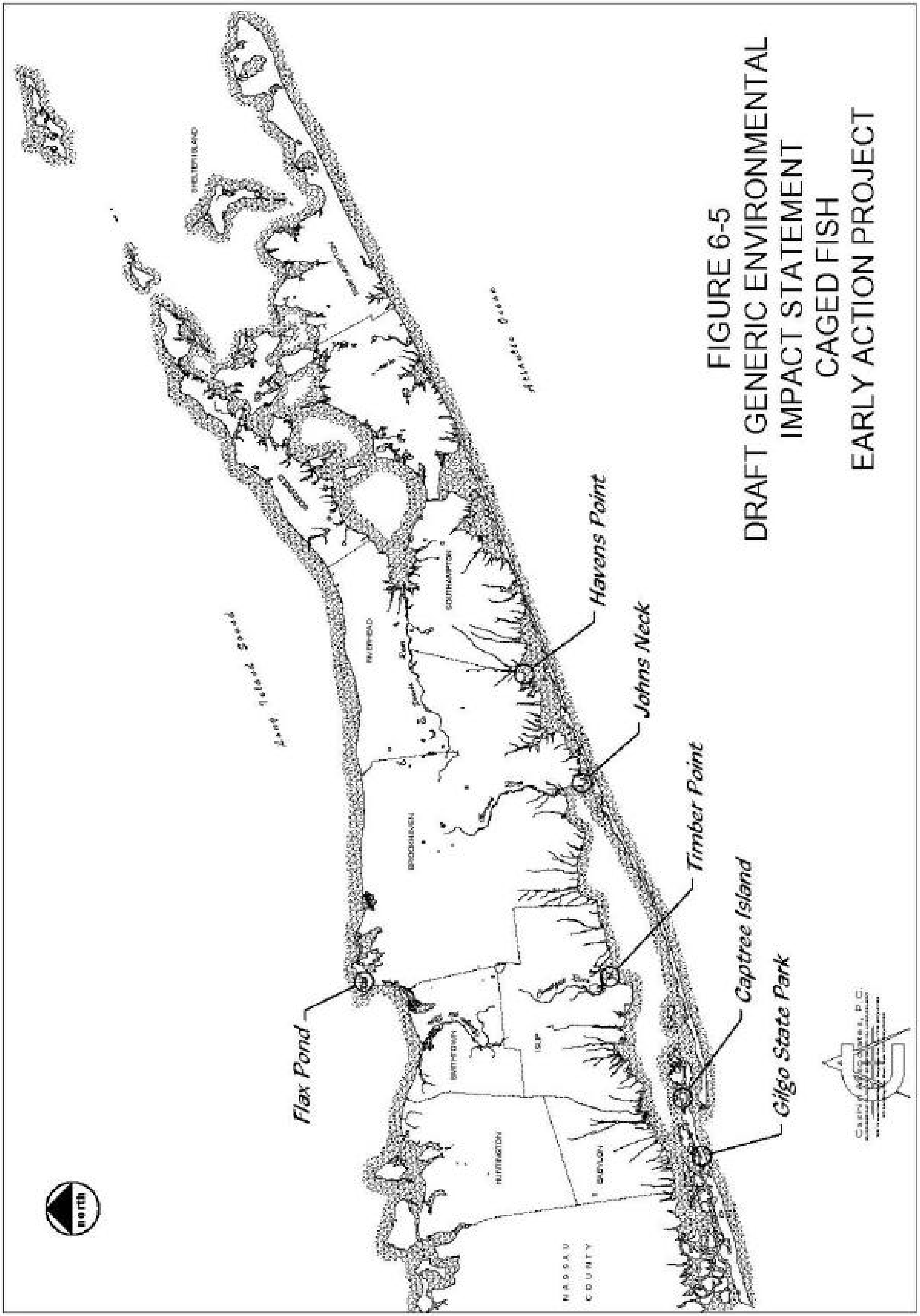
FIGURE 6-5
DRAFT GENERIC ENVIRONMENTAL
IMPACT STATEMENT
CAGED FISH
EARLY ACTION PROJECT
Havens Point
Johns Neck
Timber Point
Captree Island
Gilgo State Park
Flax Pond
north
NASSAU
COUNTY

Due to delays in securing approval to allocate funds for this project, fully monitored testing did not commence until August, and extended into the first week of September when spraying operations ceased for the season. Although good survival of caged organisms at test sites had been achieved prior to this period in preliminary studies, survival at both reference and test sites was sometimes diminished during the period of the fully monitored caging study. The study events are listed in Table 6-4.

Cages:

This study utilized a modification of Plexiglas traps designed and successfully deployed in other studies (Scott et al., 1999) for crustaceans, and simple plastic buckets with mesh inserts similar to those used in 2003 by Southampton College for fish (SCERP, 2004). Three cages for each species were deployed at each site with 14 and 20 individuals per cage for shrimp and fish, respectively. Both types of cages were tethered to bricks at the bottom and suspended from floats to allow them to move with the tides and maintain the animals at a fixed distance (three to six inches) below the surface of the water. Floats were attached so as not to interfere with direct deposition of aerosol material to the top of the mesh portions of the cages.

Test Organisms:

The sheepshead minnow (*Cyprinodon variegatus*) and the grass shrimp (*Palaemonetes pugio*) were used as representative marine fish and crustacean species in this study. The grass shrimp is a common salt marsh resident on Long Island. Furthermore, crustaceans typically are most sensitive to pesticides, thereby making them a good test organism (Clark et al., 1989). Sheepshead minnows were chosen as a fish species because they are commonly used in toxicity studies and were used in a pilot study on mosquito control applications on Long Island conducted by Southampton College in 2003 (SCERP, 2004; Wirth et al., 2001). As with the grass shrimp, sheepshead minnows are also commonly found in Long Island salt marshes. The use of young fish allowed growth rates to be measured, which provided a sublethal measure of effect not available by using adult fish as a test species. The fish were purchased from Cosper, Inc., a professional bioassay company, prior to each experiment, ensuring identical age and similar health, size, and genetic characteristics.

Table 6-4. Caged Fish Events

Experiment Dates:	Spray Dates:	Spray Type	Sites Involved:	Spray Sites:	Animals Involved:	Chemistry Performed	DO Evaluated:
7/20 – 7/26	7/20	L	Johns Neck	JN & TP	Fish	None	No
			Timber Point				No
			Old Fort Pond				No
			Havens Point				No
8/2 – 8/7	8/3	L	Timber Point	JN & TP	Fish & Shrimp	Water samples at 1,	Yes
			Johns Neck			2, 24, 48 & 96 h post-	Yes
			Havens Point			Spray; sed. Samples at	Yes
			Flax Pond			1 & 4 days post-spray	Yes
8/9 – 8/14	8/10	L	Timber Point	JN & TP	Fish & Shrimp	Water samples at pre-	Only 8/9 - 8/12
			Johns Neck			spray & 30min., 1d & 4d	Only 8/9 - 813
			Havens Point			post-spray; sed. Samples	Yes
			Flax Pond			At 1 & 4d post-spray	Yes
8/17 – 8/22	8/18	A	Johns Neck	JN	Fish & Shrimp	water & sed samples same	Yes
		(L on 8/17)	Havens Point			as on 8/3.	Yes
8/25 – 8/29	8/25	A	Johns Neck	JN	Fish & Shrimp	Water samples at pre &	Yes
			Havens Point			post 30min. & 1d & 4d post	Yes
8/31 – 9/5	9/1	L	Johns Neck	TP	Fish & Shrimp	water & sed samples same	Yes
			Timber Point			as on 8/25.	Yes
			Havens Point				Yes

Spray Type: A = Adulticide and L = Larvicide

All test animals were held and maintained either at the Flax Pond Marine Laboratory in Stony Brook (shrimp) or Southampton College's Marine Station in Southampton (fish) prior to use.

Organism Assessment:

The study plan called for both shrimp and fish to be deployed the day before a spray event, and their survival checked immediately prior to the application. Shrimp and fish survival were observed daily for the duration of each spray experiment (four or five days). Dead organisms were removed so as not to deleteriously influence the continued survival of live organisms. Length of juvenile fish was measured prior to deployment in the field and also after retrieval at the end of each experiment in order to determine relative growth rates. The shrimp that survived field exposure were brought back to the laboratory and their prey capture rates were determined in feeding trials with live brine shrimp. The ability to locate and successfully capture prey is an excellent measure of locomotive and sensory ability that has been shown to be compromised by sublethal exposure to contaminants found in urban estuaries (Perez and Wallace, 2004). Prey capture response studies were performed on a large proportion of field spray survivors following the completion of each spray experiment. If no (or few) survivors were left at a particular site, individuals used in the static tests were used instead. Large dissection bowls 20 cm in diameter were filled with one L of ultra-violet sterilized and filtered Flax Pond seawater and experiments were run for one hour. At the beginning of the experiment, five brine shrimp were placed in the center of all bowls, and every 15 minutes the number of prey consumed was observed and replenished. At the end of the hour, average prey capture rate was calculated and compared among groups.

In addition to the measurements taken on field-collected organisms, static tests were also conducted on reference shrimp exposed to water collected just after each spray event. Approximately 30 minutes following each spray event, four L of water from both spray and control sites was collected in dark bottles for static survival tests. Small dissection bowls were filled with 150 mL of water collected from each site, and shrimp that had become acclimated to laboratory conditions, were placed one per bowl in six bowls per site for the duration of each spray event. The pesticide/water solution in the bowls was replenished once daily, and at this time, shrimp survival was observed. These tests provided an independent test of the toxicity of surface waters immediately after spraying that would not be influenced by site specific water quality factors, cage failure, or loss.

Water Quality:

Data on water temperature and dissolved oxygen (DO) (as percent saturation) were collected for the duration of each experiment. WTW 340i dissolved oxygen meters with Durox probes were used to obtain continuous oxygen data in percent form. Dissolved oxygen meters monitored and recorded these variables at 30 minute intervals, and YSI Model 85 readings were taken during daily survival assessments measuring salinity, temperature, and dissolved oxygen with which to compare meter readings. A USEPA model based on actual time to death data resulting from exposure to low oxygen was used to determine whether or not diurnal hypoxia observed at many sites during the field study was in itself a significant cause of toxicity (USEPA, 2000).

Statistical Analyses:

Toxicity data from each test was analyzed by analysis of variance (ANOVA) with post hoc means tests. P-values of 0.05 or less were considered significant. To account for pre-spray mortality, survival data on day four was corrected to the percentage of organisms surviving after one day of deployment, pre-spray. This corrected survival data was then subjected to an arcsin transformation prior to application of ANOVA. When the data set was completely balanced with respect to numbers of sprayed and non-sprayed sites, it was possible to conduct a nested two-way ANOVA to evaluated spray related effects. When data from a site was lost, usually due to high (>80 percent) pre-spray mortality, one-way ANOVAs were performed.

Results:

Survival of Caged Fish and Shrimp:

Due to mortality observed frequently after deployment in the field, but before pesticide spraying occurred, post spray survival data was normalized to survival after one day in the field to better assess subsequent changes in survival due to spraying. If survival post deployment was less than 20 percent, no correction was made and the data was excluded.

- July 20 - Larvicide Spray at Timber Point and Johns Neck with Havens Point and Old Fort Pond as reference sites: This preliminary study only examined toxicity in caged fish and did not examine post deployment, pre-spray mortality. Two-way nested ANOVA revealed spray related effects (p=0.030) but site-specific tests showed that Johns Neck was the only site showing significantly reduced survival as compared to all other sites including Timber Point.

- August 3 - Larvicide Spray at Johns Neck and Timber Point with Flax Pond and Havens Point as reference sites: Two-way nested ANOVA of the fish data did not reveal significant spray related differences (p= 0.059), although mortality in the sprayed sites tended to be higher. Shrimp survival at Timber Point was less than 20 percent after their first night in the field (assessed a few hours after spraying); therefore these data were not included in the statistical analysis, or in the figure showing corrected data. One-way ANOVA showed significant differences between shrimp mortality at the other three sites, with mortality at Johns Neck significantly higher than that observed at Flax Pond.

- August 10 - Larvicide Spray at Johns Neck and Timber Point with Flax Pond and Havens Point as reference sites: Due to the poor survival at Timber Point during the August 3 spray study, cages were moved to deeper water in this and subsequent experiments at that site. Two-way nested ANOVA showed no significant spray related effects (p=0.09), although there were significant differences between sites when compared to Havens Point, showing increased mortality with respect to Flax Pond or Timber Point. Two-way nested ANOVA of the shrimp data indicated a significant increase in mortality at sprayed sites (p=0.050). However, individual site comparisons showed increased mortality at Havens Point as compared to Flax Pond and Timber Point, and at Johns Neck as compared to Timber Point and Flax Pond.

- August 18 - Adulticide Spray at Johns Neck with Havens Point as a reference site: One-way ANOVA did not show significant differences between mortality at the sprayed and non-sprayed sites for fish, although shrimp mortality was significantly elevated at Johns Neck as compared to Havens Point (p=0.0005). It should be noted that larvicide was sprayed at Johns Neck just after animal cages were deployed and about 24 hours before adulticide was sprayed at these sites.

- August 25 - Adulticide Spray at Johns Neck with Havens Point as a reference site: Due to the continued high mortality in the ditch at Johns Neck prior to spraying, cages were also deployed in additional locations at Johns Neck prior to this spray event. In addition to placing cages six inches below the surface in the ditch as had been done previously, cages of fish and shrimp were also deployed in the main channel at the entrance to this ditch, and cages of fish were also deployed right at the surface in the ditch at Johns Neck and also at Havens Point. For both fish and shrimp, survival was excellent in the cages placed in the main channel at Johns Neck. Significant mortality was observed at the Johns Neck ditch site as compared to the Johns Neck channel or Havens Point sites, for both species. Significant mortality was also observed at Havens Point as compared to the Johns Neck channel site for fish. It should be noted that in this experiment, organisms were deployed the same day as the spray, so there was no opportunity to assess pre-spray mortality.

- September 1 - Larvicide Spray at Timber Point with Havens Point and Johns Neck Channel used as reference sites: Only Timber Point was sprayed on this date, but cages were deployed both at Havens Point and Johns Neck for comparison. For fish, significantly greater mortality was observed at Johns Neck and at Havens Point as compared to Timber Point. No significant differences were observed in shrimp mortality, although it should be noted that shrimp were not deployed at the Johns Neck ditch site during this event.

Diel Patterns of Dissolved Oxygen and Temperature:

Data on the patterns of dissolved oxygen (DO) measured by probes deployed on one cage at each site, were collected. At the time of sampling, DO, temperature, and salinity were also measured using a YSI meter and probe. In general, the field deployed recording DO/temperature meters agreed well with the YSI probes.

Significant diel variation was observed at all sites. Often, DO crashed twice a day at Johns Neck. Periods of low DO were most pronounced in the ditch at Johns Neck and at the first site at Timber Point (used in the August 4 deployment). As both the severity of hypoxia and its duration are important determinants of low DO toxicity, a time to death approach developed by the USEPA (USEPA, 2000) was used to estimate when expected toxicity resulting solely from periodic DO fluctuations would occur. With the help of Glen Thursby (one of the principal developers of the USEPA approach), this approach was applied to data obtained for adult *P. pugio* to determine on which days and at which sites toxicity due solely to DO, could be expected. Two criteria were examined:

- hypoxia likely to exceed the LC_{50} for shrimp (less than five percent saturation for one or more hours, or less than seven percent saturation for four or more hours during any 24 hour period)

- hypoxia likely to exceed the no observable effects threshold for shrimp (less than nine percent saturation for two or more hours during any 24 hour period).

Unfortunately, time to death data, in response to controlled hypoxic conditions, were not available for the sheepshead minnows. The juvenile fish used in this study appeared to be less sensitive to low DO (there were several cases where all the shrimp died, but reasonable fish survival was observed). Therefore, the use of critical values available for the shrimp seemed appropriate. Using these criteria, toxicity due to low DO alone could have been expected to contribute to mortality in fish:

- at Johns Neck and Timber Point during the August 3 spray study;

- at Havens Point and Johns Neck ditch during the August 18 and August 25 spray studies;

- on some days (September 1, September 3, and September 4 at Havens Point; and

- on all days at the Johns Neck ditch location during the last spray study.

Laboratory Static Renewal Shrimp Toxicity Studies:

Four day static renewal toxicity tests conducted in the laboratory on grass shrimp exposed to water collected at the depth of the cages 30 minutes after spraying, indicated no toxicity associated with exposure to this water from the spray sites.

Growth in Caged Fish:

Length measurements taken on fish prior to and after field deployments were made. Growth in these fish was relatively small with large variability between groups. As there were no obvious trends in fish growth, statistical analysis could not be conducted.

Prey Capture Studies:

Shrimp surviving field deployment were also tested for their ability to capture and consume live adult brine shrimp in the laboratory. In cases where there were insufficient numbers of surviving shrimp from the field deployment, shrimp exposed to water collected from the field in the static renewal toxicity tests were assessed. No significant differences between sites were observed except during the September 1 larvicide spray at Timber Point. In this case, shrimp deployed at Johns Neck showed significantly lower prey capture ability when compared to shrimp deployed at either Havens or Timber Point ($p<0.002$).

Discussion:

The original plan for this study called for all field work to be conducted prior to the beginning of August to avoid anticipated low DO events that are more prevalent during the hottest period of the summer. Unfortunately, due to many delays in obtaining permission to conduct the study, this was not possible. Preliminary data on caged fish and shrimp survival at all sites showed good survival during July. However, by the time the fully replicated study was performed, this was not the case. Periodic low DO was prevalent at the ditch site in Johns Neck and at the ditch site in Timber Point used during the August 3 spray event (the cages were moved into more open water for subsequent spray events). Later in August, and for the early September spray, low DO was also a problem at the Havens Point reference site. These problems with low DO compromised the ability to detect toxicity that may have been due to pesticide exposure.

During this study, larvicide spraying was tested on four occasions (July 20, August 3, August 10, and September 2). During the first spray, reduced fish survival was observed at Johns Neck, as compared to Timber Point, which was also sprayed, as well as Havens Point and Old Fort Pond, reference sites. Unfortunately, no DO measurements were taken, so it is not known whether or not low DO could have been a factor at Johns Neck. The reduced survival for shrimp observed at Johns Neck and Timber Point, as compared to the reference sites at Havens Point and Flax Pond during the larvicide spray on August 3 could be attributed to low DO alone. During the August 10 spray, reduced survival was observed both at Johns Neck and at Havens Point, even though low DO should not have been a problem. During the September 1 spray, Timber Point, the only site sprayed, showed the best survival of all the sites evaluated at that time. Finally, in the static renewal studies conducted in the laboratory using water collected 30 minutes post spray at each site, excellent survival was observed in shrimp exposed to water from the spray sites.

Adulticide applications were only tested on two occasions, one on August 18, and one on August 25, both at Johns Neck only. It had been anticipated that adulticide spray impacts at approved sites in Gilgo State Park could be tested, but no aerial applications occurred near there. Unfortunately, low DO was persistent enough at both the Johns Neck ditch site and at Havens Point during these spray events to contribute to mortality. In the first study, enhanced fish mortality was observed only at Havens Point, whereas enhanced shrimp mortality was observed at Johns Neck. In the second adulticide spray, organisms were also caged in the creek at Johns Neck. There was no evidence of increased mortality at the Johns Neck creek site, while significant mortality was again observed at Havens Point. Finally, no evidence of reduced survival was observed in static tests with shrimp, which was performed in the laboratory following these adulticide applications.

Cashin Associates (2005b) contains a fuller discussion of this part of the experiment.

6.3. Impacts to Caged Organisms from Vector Control Pesticides Experiment, Part 2: Pesticide Aquatic Fate and Transport

This portion of the experiment is based on Cashin Associates (2005c), unless otherwise explicitly referenced.

Chemical analyses were conducted by a team including SCDHS (through the PEHL), USGS, and the Bruce Brownawell laboratory, Marine Sciences Research Center, Stony Brook University. Funding was provided, as discussed above, through the County Legislature. The Brownawell laboratory was funded partially through a direct subcontract with CA and partially through CA' agreement with the State University Research Foundation. USGS was funded through a cooperative agreement with SCDHS.

The chemical analyses included the following elements:

- SCDHS conducted broad-spectrum water sampling at the time of the site pre-testing, to ensure non-SCVC related chemicals or substances are not present at sufficient concentrations to compromise the experiment.
- 60 aqueous samples were analyzed by the Brownawell laboratory.
- 20 aqueous samples were analyzed by USGS.

Sampling included pre- and post-spray water (surface and subsurface), as well as sediment sampling, together with 96-hour sample acquisition. A more detailed time series event for one adulticide and one larvicide event was also conducted (see Table 6-4). The Brownawell laboratory analyzed a total of 60 sediment samples, and USGS analyzed 10 sediment samples. Organisms (40 samples) at the test and control sites were also analyzed for pesticide residues by the Brownawell lab. Chemical sampling was coordinated and carried out under the direction of Steven Terracciano of USGS, and the analyses were directed by Dr. Brownawell.

An intercalibration study between USGS, SCDHS, and MSRC to compare water analysis methods, determine sample variability, and precision of analysis was also conducted. This work was organized by Ken Hill of PEHL. The intercalibration study allows the results obtained in this experiment to be compared to the much larger data set obtained by USGS on levels of pesticides in surface waters after spray events.

It is clear that the adulticide, resmethrin, is not persistent, as it was never detected more than two hours after the applications, despite achieving very low detection limits (Table 6-5). The Brownawell laboratory was able to achieve very low detection limits through the use of HPLCtime of flight-MS analyses. The adulticide synergist, PBO, is more persistent (Table 6-6). Methoprene was also slightly persistent, but is also is difficult to detect in the water column after aerial applications (Table 6-7). None of the target compounds were detected at control sites, nor did SCDHS testing find any organic compounds in the pre-application water samples.

Table 6-5. Resmethrin Data Summary

Sample Type	**Result**	**Notes**
Water Column	320 ng/l	0.5 post-spray, water interface maximum concentration
Water Column	38 ng/	2 hrs post-spray, subsurface maximum concentration below interface
Water Column	2 hrs	Last detection post-spray
Sediment	ND	All samples
Method Detection Limit (MDL), USB	500 pg/l	
MDL, USGS	5 ng/l	
Instrument Detection Limit (IDL), USB	230 pg/l	

Table 6-6. PBO Data Summary

Sample Type	**Result**	**Notes**
Water Column	59.8 ug/l	0.5 post-spray, water interface maximum concentration
Water Column	2.9 ug/l	2 hrs post-spray, subsurface maximum concentration below interface
Water Column	48 hrs	Last detection post-spray
Sediment	ND	All samples
MDL, USB	500 pg/l	
MDL, USGS	5 ng/l	
IDL, USB	25 pg/l	

Table 6-7. Methoprene Data Summary

Sample Type	**Result**	**Notes**
Water Column	3.3 ug/l	0.5 post-spray, water interface maximum concentration
Water Column	> 2.5 ug/l	0.5 hrs post-spray, subsurface maximum concentration below interface
Water Column	44 hrs	Last detection post-spray
Sediment	3-60 ng/g	Samples collected within 1 week
MDL, USB	500 pg/l	
MDL, USGS	5 ng/l	
IDL, USB	90 pg/l	

Methoprene could sometimes be measured at relatively high concentrations (at or in excess of one ppb) immediately following an application in waters surrounding a target site. However, these concentrations were not sustained with time, and within two days of an application samples no longer contained concentrations greater than 10 ppt (see Figure 6-6). Some very low concentrations of methoprene were detectable, on occasion, more than one week after an application (data presented in the graph as both pre-spray detections, and detections at 144 hours post-spray).

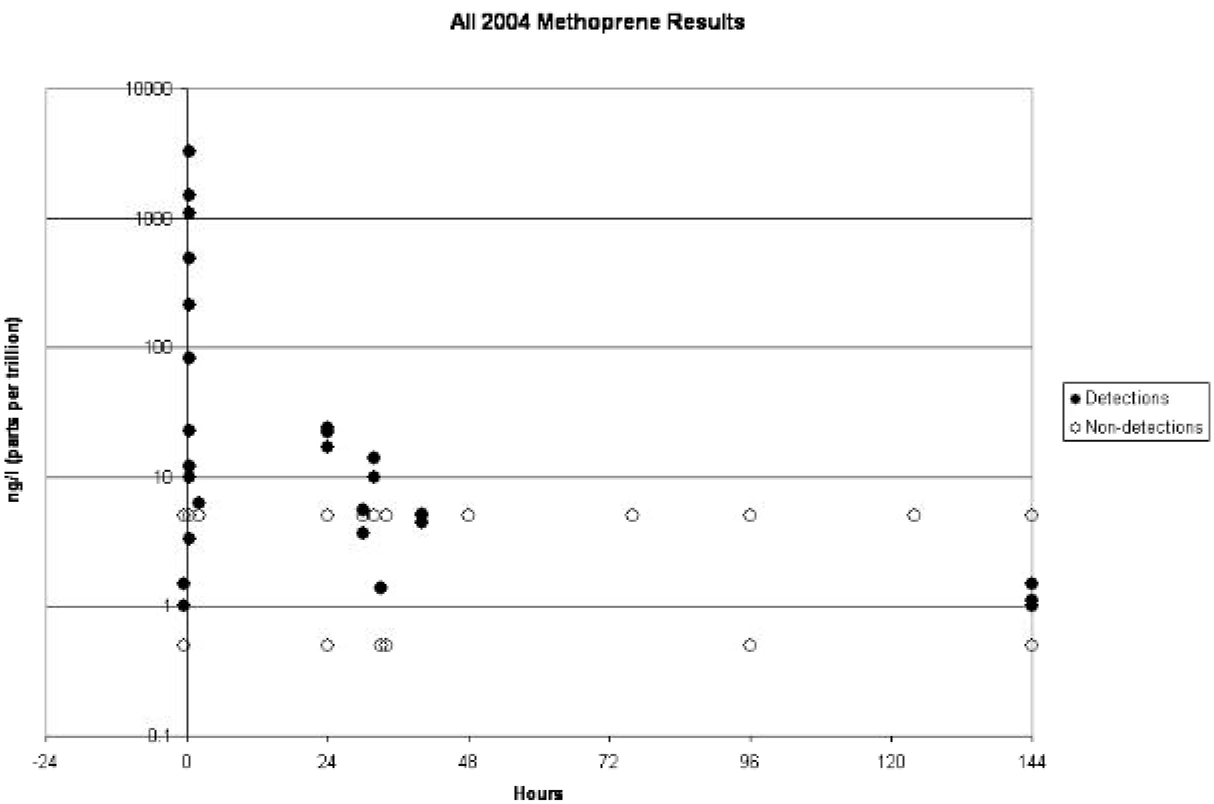

Figure 6-6. Time series of all methoprene data at Timber Point and Johns Neck ("144 hour" data include later data points)

Immediately following an application, it was possible to measure resmethrin in aqueous samples. The greatest concentrations were measured immediately after the application. The concentrations fell off rapidly with time, and by pre-dawn following an evening application resmethrin was not detectable in the water column. PBO was found at much higher concentrations, even accounting for its greater relative concentration in the applied pesticide mixture, immediately after the applications. However, its concentration, too, was reduced from maximum measurements (less than 10 ppb) to 100 ppt or less within nine or 10 hours, and it was very difficult to measure even single ppt concentrations of PBO after two days or more following the two applications (see Figure 6-7).

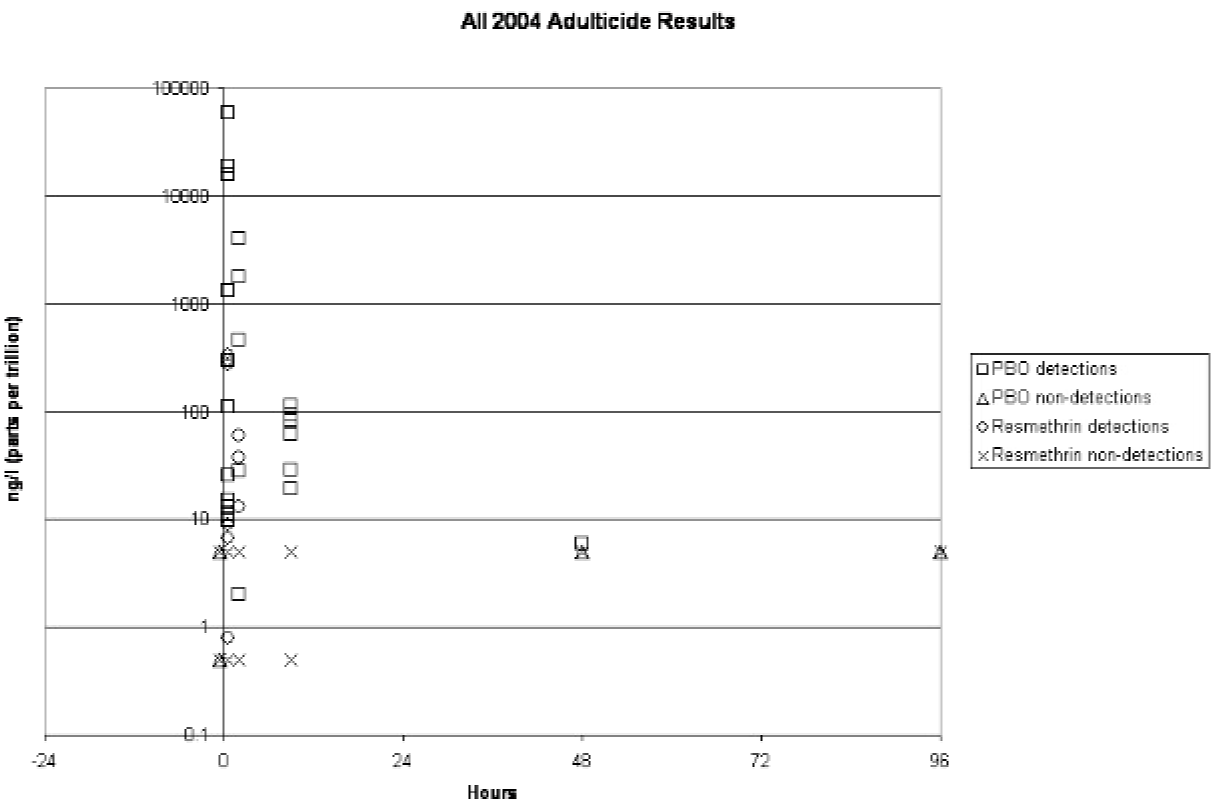

Figure 6-7. Time series of all resmethrin data from Johns Neck

Sediment sampling did not find any of the adulticide compounds. Methoprene was detected in sediments at sites where applications occurred. Methoprene was not detected in any samples from sites where it had never been applied (Havens Point and Flax Pond).

From these data sets, conclusions regarding the fate and transport of these compounds in mosquito ditches can be drawn. Resmethrin appears to be enriched in the water interface. This would suggest that it is associated with particulate matter, which might explain why it was difficult for USGS to detect in subsurface waters (USGS filters all samples). However, the Stony Brook team did not filter its samples, and also had difficulty detecting the compound. Resmethrin was also not detectable in sediment samples, as might be expected if it were adhering to particulate matter (which might settle out of the water column). However, even at the water interface immediately after pesticide application, resmethrin is depleted compared to PBO, the synergist. In the Scourge formulation, the ratio of resmethrin to PBO is 1:3. At the interface, the ratio was from 1:50 to 1 to several hundreds. One subsurface sample ratio was as high as 1:14, but it is clear that resmethrin concentrations decreased much faster than PBO concentrations did. This suggests that resmethrin is rapidly degrading in the environment – although the mechanism for such decay is not yet determined.

Methoprene is intended to sink through the water column, and so it is not surprising that it was not detected more than a few hours following applications. Thus, it is also not surprising it was detectable in sediments. What was unusual was that the concentration of methoprene in the sediments did not increase with time. That suggests, given a one week application cycle for many sites, including most of those tested in this experiment, that the methoprene must have a half-life considerably shorter than one week.

These data show that the chemicals used for mosquito control tend to have very short persistence in the water column, even when measurements are made at the high part per quadrillion (pg/l) level. Methoprene persists in sediments for some time, but also degrades at such a rate that accumulation in the sediments did not occur despite weekly re-applications.

More complete discussions of the Brownawell (Cashin Associates, 2005c) and USGS (CA-USGS, 2005) results are available. The USGS reports include a summary of all its work on mosquito control pesticides testing in Suffolk County for 2001 through 2004.

Proficiency results were reported in February, 2006. Two samples with equivalent concentrations of analytes of interest were delivered to each of the three laboratories that have analyzed for the pesticides during the Long-Term Plan. One was spiked with "low" concentrations of the analytes of interest; the other was spiked with higher concentrations.

Tables 6-8 and 6-9 report the results. It is clear that all three of the laboratories performed very well on nearly all analytes at the two concentrations; the sole results that were not within 25 percent of the true concentration were the USGS results for the higher concentrations of resmethrin and PBO, both of which fell just outside of that subjective range. The mean differences between results and true values ranged from approximately eight percent to just over 20 percent, which suggests that the values reported for field samples are most probably close to the actual concentration that existed in the water samples.

Table 6-8. NSI Low Concentration Proficiency Results

Analyte	True Value (ng/l)	SUNY Result (ng/l)	Percent Agreement	USGS Result (ng/l)	Percent Agreement	PEHL Result (ng/l)	Percent Agreement
Resmethrin	93.4	81	86.7	80.5	86.2	80	85.7
PBO	106	120	113.2	97.3	91.8	110	103.8
Sumithrin	80.4	72	89.6	76.8	95.5	90	111.9
Malathion	138	120	87.0	154	111.6	150	108.7
Methoprene	174	150	86.2	170	97.7	210	120.7
Mean Percent Agreement			92.5		96.6		106.2
Mean Percent Deviation			12.7		8.1		11.9

Table 6-9. NSI High Concentration Proficiency Results

Analyte	True Value (ng/l)	SUNY Result (ng/l)	Percent Agreement	USGS Result (ng/l)	Percent Agreement	PEHL Result (ng/l)	Percent Agreement
Resmethrin	467	420	89.9	310	66.4	440	94.2
PBO	531	640	120.5	390	73.4	600	113.0
Sumithrin	402	340	84.6	329	81.8	430	107.0
Malathion	688	630	91.6	772	112.2	860	125.0
Methoprene	872	740	84.9	768	88.1	860	98.6
Mean Percent Agreement			94.3		84.4		107.6
Mean Percent Deviation			13.9		20.5		10.6

6.4. Impacts to Caged Organisms from Vector Control Pesticides Experiment, Part 3: Benthic Sampling for Impacts from Seasonal Exposure to Pesticides

Material in this section, unless otherwise noted, is from Barnes (2005).

Five locations were sampled for benthic invertebrate community impacts from pesticide use over the course of the season. Two sites were exposed to pesticides in 2004:

- Johns Neck (*Bti*, methoprene, resmethrin)
- Timber Point (*Bti*, methoprene)

These will be referred to as treated sites. Three sites were selected that were not exposed to any larvicides or adulticides in 2004:

- Captree Island (West)
- Havens Point
- Flax Pond

This work was accomplished by Robin Barnes from the McElroy laboratory, Marine Sciences Research Center, Stony Brook University. Funding was provided by the County Legislature. Ms. Barnes was funded by the CA agreement with the State Research Foundation; and Drs. McElroy and Cerrato, who served as project supervisors, were funded through direct subcontracts with CA.

6.4.1. Sample Collection and Processing

October 15 to October 19, 2004, benthic samples were collected from Havens Point, Johns Neck, Timber Point, and Captree Island to study pesticide effects on community structure after pesticide applications had been completed for the season. Three core samples were collected at each of the sites in close proximity to identified cage locations during the caging study (note that the Captree Island site, although scouted for potential use in the Caged Fish study, was not actually used in the main study, discussed above). The top 10 cm of each core was extracted and run through a 500 µm sieve to exclude most of the detritus. The entire sample remaining in each sieve was placed in several four ounce jars per core and preserved with 10 percent formalin. Approximately six months after the core samples were collected; they were transferred to a mixture of 70 percent ethanol and rose Bengal in order to stain the biological organisms present in the samples. Due to the large volume of material collected from each core, only one jar worth of material from each core was analyzed. Analysis included using a dissection microscope to identify all species found, down to the lowest taxa possible and count the number of each different organism. Worms were only counted if heads were found. Photographs of each type of organism collected were also taken to allow for confirmation of species identifications and for future species analyses by others.

6.4.2. Data Analysis

The PERMANOVA program was used to carry out nested, nonparametric, multivariate analysis of variance on the benthic community data (Anderson, 2001). Four groups of species data were analyzed:

1) all species

2) only marine species (several terrestrial organisms were excluded)

3) only crustacean species

4) all arthropod species.

Results obtained from the program tested whether the treated sites were significantly different in composition and abundance from control sites, using $p < 0.05$ as a test of significance. In order to determine site-by-site differences, pairwise comparisons were made using PERMANOVA. Due to the low number of replicates obtained (and thus the low number of possible permutations allowable by the program), the Monte Carlo asymptotic p-value, rather than the permutation p-value, was used for determining significance.

Twenty-six particular organism identifications were made, some to the species level (six organisms), some to the genus level (five organisms), and others to family level. In all cases, identifications were to the lowest taxa possible. Organism abundance and diversity at larvicide sites were not found to be significantly different from those observed at the control site, regardless of what groups of species were tested.

There were significant differences in species abundance and diversity in the pairwise comparisons. For the all species included analysis, the following sites were found to be significantly different from each other (treated sites are specially identified):

- Havens Point and Captree Island
- Havens Point and Johns Neck (treated site)
- Havens Point and Timber Point (treated site)

When only marine organisms were considered, Havens Point was still different from Timber Point (treated site). The analysis of Johns Neck (treated site) and Havens Point failed to meet the test of statistical significance by only the smallest margin (p = 0.0545).

The analysis of crustacean species only, found no statistically significant differences, although the comparison of Havens Point and Captree Island (p = 0.0559) and Captree Island and Johns Neck (treated site) (p=0.0543) nearly met the criterion.

The analysis of all Arthropods, found two pairs that were statistically significantly different:

- Havens Point and Captree Island
- Havens Point and Johns Neck (treated site)

The pairing of Havens Point and Timber Point (treated site) (p = 0.0528) nearly met the criterion for significant difference.

It is clear from the above pairings that the benthic community at Havens Point was different from most other sites, using several different criteria to make comparisons. Havens Point sediments tended to be sandier than the sediments from the other three sites. It is well-known that substrate differences can lead to community structure and biomass differences for benthic invertebrates (Cerrato et al., 1989). Therefore, the benthic population differences found in the pairwise comparisons may relate closer to environmental differences, than whether pesticides were applied or not in 2004. This conclusion is supported by the lack of differences when the communities of the two pairs of treated sites were compared to those at the two untreated sites.

6.5. Wertheim National Wildlife Refuge-Seatuck National Wildlife Refuge Marsh History Determination Project

Unless otherwise noted, the material in this section is drawn from Cashin Associates (2006b).

6.5.1. Background

Salt marshes accrete materials in order to maintain their elevation against sea level rise. A portion of this accreted material is inorganic, comprised of sediments washed either from