Scope of Study for 4-Poster SLN Permit: Amendment Request for Objective II

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Objective & Introduction: The scope of study outlines the experimental portions of the Special Local Need (SLN) permit for the 4-Poster acaricide delivery system on Fire Island and Shelter Island (Long Island, NY). The 4-Poster study addresses technical concerns held by the New York State Department of Environmental Conservation (DEC) and Department of Health (DOH) regarding 4-Poster registration and use in New York State. Objective II from the scope of study was designed to investigate permethrin residues in and on white-tailed deer from 4-Poster treatment areas. This original objective was devised and approved in 2007 and then revised and approved in 2008. Laboratory analytical detection of permethrin residues within the muscle of 3 of 16 sampled deer on Shelter Island during 2008 has prompted this second amendment of Objective II to be submitted during July 2009 for review by the NYSDEC and NYSDOH.

Overview of Technical Concerns: The concerns of NYSDEC and DOH broadly fall into three general categories:

I. Human and wildlife-associated risks due to change in deer movement and behavior. Voiced concerns include:
   - potential impact on vegetation near the deployed stations in both natural and residential settings
   - possible increase in deer road crossing leading to more vehicle collisions
   - increased contact and potential disease transmission between deer
   - changes in deer mortality due to feeding or reduced tick pressure
   - use of feeding stations by non-target wildlife.

II. Possible increased human exposure to permethrin via handling and consuming treated deer. Concerns expressed are dermal exposure via the deer hide during handling as well as the possibility of permethrin accumulation in deer tissues that would be orally ingested by consumers of venison.

III. Efficacy of the 4-Poster system in controlling tick densities in human inhabited and visited areas. The 4-Poster label dictates that stations must not be deployed within 100 yards of anywhere a child may be present without adult supervision. It has been agreed that a reduction in tick densities can serve as the best proxy for a reduction in the likelihood of human disease transmission.

**** The original Objective II was devised and approved in the original Scope of Study during 2007.***

Objective II. Investigation of Permethrin Residues in and on Deer from Treated Areas.

There are no public deer check stations on Shelter Island, so we will work with Town staff to collect tissue samples and swabs from deer harvested under Bonus Deer Permits issued by DEC. If the entire developed area on Shelter Island is treated with 4-Poster stations, these deer can be sampled outside of the 2,400-acre study sites. The heart, liver, and a sample of muscle of harvested deer will be stored in glass containers, frozen, and tested for permethrin residues at the Animal Health Diagnostic Laboratory at Cornell using a mass spectroscopy technique. Deer from 4-Poster treatment areas will be collected in collaboration with the Town of Shelter Island Deer Management Program during November and December. Deer hides will be wiped with cotton swabs along both sides of the head and neck in areas likely to contact 4-Poster rollers. We will focus on tagged deer that we know have contacted 4-Poster stations based on the IR-camera photos. These samples will be tested for permethrin by a chromatography technique (Miller et al. 1983). The sample size will be approximately 45 animals for the entire duration of the study (about 15 deer per year). This number includes a sample of 5-10 deer from the control site in North Haven so that we can evaluate background permethrin levels (potential contact from lawn spraying).

**** The first amendment to Objective II was completed and approved during 2008.***

Objective II. Proposed Revisions for Permethrin Residue Investigations

To evaluate potential human exposure to permethrin through contact with deer hides and consumption of deer meat, tissues and pelage will be sampled from 30 deer (10 per study year) on Shelter Island. Check stations will be established during October – December to obtain samples from hunter harvested deer during the regular and nuisance hunting seasons. Although deer harvested from any location on Shelter Island (7,000 acres) will be sampled, tagged deer observed using 4-Poster devices in IR-camera photos will be primarily targeted for sampling.

To evaluate base line permethrin levels or potential permethrin accumulation in deer from broadcast lawn spraying, tissues and pelage from 15 deer (5 per study year) will be sampled in the control site of North Haven. These control samples will provide information that is representative of potential human exposure to permethrin preceding 4-Poster use. Individual hunters will be contacted or check stations will be established during October – December to obtain samples from harvested deer on North Haven.

From each deer, 1 liver and 1 muscle sample will be collected for analysis. These samples will be stored in glass containers, frozen, and tested for permethrin residues at the Animal Health Diagnostic Laboratory at Cornell using a mass spectroscopy technique. Deer pelage in areas likely to contact 4-Poster rollers (both sides of the head

and neck) will be wiped with 1 cotton swab. Swab samples will be analyzed for permethrin by a chromatography technique (Miller et al. 1983).

**** The second amendment to Objective II was completed and submitted for approval in July 2009.***

Objective II. Investigation of Permethrin Residues in and on Deer from Treated Areas.

Sampling Objective

Objective II is designed to address concerns of potential human exposure to permethrin via handling and consuming deer from treatment areas. Residue sampling is conducted to detect permethrin residues on deer coats and within deer tissues as well as identify potential risks associated with hunting and consuming venison from areas where 4-Poster technology is used. Similar to sampling conducted during 2008, the sampling conducted during 2009 and 2010 will continue to address the aforementioned sampling objectives related to potential human exposure. Residue sampling will be reassessed upon completion of the 2009 fall sampling season. The Cornell Animal Health Diagnostic Center will continue to conduct all permethrin residue analyses for the 4-Poster Deer and Tick Control Study.

Sampling Plan

Thirteen deer on Shelter Island and 6 deer on North Haven will be collected for permethrin residue sampling during 2009. Permethrin residue sampling will continue during 2010 and sample collection is anticipated from 10 deer on Shelter Island and 5 on North Haven. The deer sampled during the regular fall deer harvest seasons of 2009 and 2010 will be prioritized based on verified 4-Poster device use. Verified device use will be determined for each deer through positive permethrin detections on coat swab samples or infrared-triggered camera detection methods. Sample analysis based on positive coat swab detections will require submission and analysis of swab samples prior to further analysis of muscle and organ samples.

Sampling will include collection of coat swab, muscle tissue, and organ samples from deer on Shelter Island and North Haven. Two muscles (1 neck and 1 hind quarter), 1 liver, and 1 coat swab (4 samples total/deer) will be sampled from each deer submitted for analysis. Sample collection methods will be consistent between Shelter Island and North Haven. Information about each deer sampled will be collected including date and location of harvest/mortality, sex, approximate age, and any sample-related comments or observations.

Permethrin residue sample collection from deer on Shelter Island and North Haven allows for comparisons between residues within the 4-Poster treatment area and the North Haven control area. Samples collected from Shelter Island will be representative of the residues hunters may potentially be exposed to during deer harvest, meat processing, and

meat consumption for those deer that use the 4-Poster devices. Deer samples obtained from North Haven, the control area, will provide base-line permethrin levels or potential permethrin accumulation in deer from broadcast lawn spraying. Similar to the 4-Poster tick control technology, lawn spraying often uses the common insecticide, permethrin. Deer samples from the control site are representative of potential human exposure to permethrin as a result of handling and consuming deer prior to 4-Poster use on Shelter Island.

Three deer of the 13 deer to be sampled on Shelter Island during 2009 will be sampled prior to the fall deer harvest season and regular fall study sampling. These deer will be positively identified using 4-Poster devices based on infrared-triggered camera detections. The samples will be collected prior to August 10, 2009 and submitted to Cornell Animal Health Diagnostic Center (AHDC) upon completion. A minimum 30-day time frame is required by the laboratory for analysis and report completion; the time may be substantially longer depending on laboratory work load. Analysis results are expected prior to the 2009 fall deer harvest season. Pre-season (summer) tissue and coat sampling should be considered ancillary to the fall season sampling. Seasonal differences in deer pelage and feeding habits affect exposure to 4-Poster Tickicide and use of 4-Poster devices. Consequently, residue data from samples collected during summer may not be entirely representative of those collected during the regular fall harvest season.

Regular fall study sampling will begin October 1, 2009. Ten deer from Shelter Island and 6 deer from North Haven will be sampled during the regular fall permethrin residue sampling during 2009. Individual hunters will be contacted during October – December 2009 to obtain samples from harvested deer on Shelter Island and North Haven during the regular fall deer harvest season. The Town of Shelter Island has been asked to assist with sampling through the submission of the first deer harvested each month (October – December) from 5 designated nuisance hunting properties. All samples will be collected by mid-December, prior to the seasonal removal of 4-Poster devices on Shelter Island. Samples will be submitted to Cornell AHDC at the same time and only upon completion of all 2009 permethrin residue sampling. The exact submission date will depend on the AHDC receiving department's holiday schedule but is expected to be no later than December 31, 2009.

Sampling Procedures

Harvest and Field Dressing

Hunters will be asked to contact Cornell as soon as possible post-harvest to provide an ungutted and unskinned deer for sampling purposes. Samples collected from hunter-harvested deer will be collected within less than 12-24 hours post-harvest by Cornell staff. If sampling is not possible directly upon harvest, the hunter will be asked to contact Cornell staff immediately for further direction. The hunter will then be asked to carefully gut but not to skin his/her deer. The hunter will be directed to store the deer in a cool location and store the liver refrigerated, in a non-plastic container. A non-plastic

container is requested for storage because plastic could contribute to permethrin leaching from the samples prior to analysis. Cornell staff will skin and collect all remaining samples from deer that are gutted by the hunter. New nitrile gloves will be worn by Cornell staff when collecting samples and hunters will be asked to wear protective gloves when they are responsible for gutting their deer. Deer samples will be prioritized for analysis based on known conditions of sampling (handling and collection solely by Cornell will be priorities for analysis) as well as availability of verified 4-Poster use indicators (positive coat swabs and/or photo documentation). Detailed records will be kept for each deer sampled and all applicable information will be summarized and available for review.

Liver Collection

Liver samples (100-grams) will be obtained by Cornell staff using a new disposable, sterile scalpel and new nitrile gloves. The liver samples will be wrapped in aluminum foil, sealed in 8 oz. glass jars, and frozen. Gloves will be worn during the liver sample collection to prevent cross-contamination.

The Cornell AHDC analysts will dissect a section of liver from the inner portion of the 100-gram sample. To minimize sample contamination, precautions will be taken by the analysts to ensure no contact occurs between the inner section and the outside of the samples. Analysis will be performed by GC/MS-MS using a minimum detection limit (MDL) of 10 parts per billion.

Hide Swab Collection

Deer coat swabs will be obtained using wipes and a protocol provided by the NYSDEC Division of Solid and Hazardous Materials Laboratory (P. Furdyna, September 2008). Wipes are prepared by exhaustive soxhlet extraction with isopropanol. Each cleaned wipe is saturated with isopropanol and packaged into an I-Chem Certified 300 series four-ounce amber, Teflon-lined screw-topped sealed glass jar. Using a disposable paper template, a 200 cm^2 (4”x8”) area of fur on both lateral side of each deer’s neck (behind the ear) will be swabbed with 1 swab in a movement with and against the grain of the hair. Swab samples will be immediately placed in sealed glass jars and stored frozen until laboratory analysis. Cornell staff will be collecting these samples wearing new, nitrile gloves during swab sample collection and gloves will be changed between each collection.

The permethrin residue analysis of coat swabs will be performed by the Cornell AHDC by gas chromatography/mass spectrometry (GC/MS-MS) using a certified reference standard of permethrin to establish retention time and response (MDL 0.010 mcg/swab). A control swab (unopened wipe) will be tested by the AHDC as an interference control check.

Skinning and Muscle Collection

Prior to muscle sample collection, the entire animal will be skinned, posterior to anterior by Cornell staff. One disposable, sterile scalpel will be used to make the initial incisions in the skin. A new, disposable, sterile scalpel will be used to assist the process of peeling the skin away from the body of the animal. As the skin is peeled from posterior toward anterior, the underlying muscle is exposed for further sample collection. Caution will be taken to avoid contact between the muscle and the scalpel that was used to assist the final stages of the skinning process.

Muscle samples (100-grams) will be obtained by Cornell staff from the neck and hind quarter of each sampled deer. A new, disposable, sterile scalpel will be used to remove a 100-gram section of neck muscle from both lateral sides of a deer being sampled. The 2, 100-gram lateral neck muscle samples will be stored separate until being processed by the AHDC. One 100-gram hind quarter muscle will be removed from either 1 of the lateral sides of a sampled deer. The muscle samples will be wrapped in aluminum foil, sealed in 8 oz. glass jars, and frozen. Gloves will be changed between skin removal and muscle collection to prevent cross-contamination.

The Cornell AHDC analysts will dissect a section of muscle from the inner portion (core) for each muscle sample. To minimize sample contamination, precautions will be taken by the laboratory to ensure no contact occurs between the core and the outside of the sample. For each set of neck muscle samples per deer, the 2, 100-gram lateral neck muscle samples will be cored; the 2 core sections will be combined and homogenized into 1 sample by AHDC analysts. Analysis will be performed by GC/MS-MS using a minimum detection limit (MDL) of 10 parts per billion. A core section from each hind quarter muscle sample will be collected by the AHDC analysts and used for further analysis. No homogenization will occur and analysis will be performed by GC/MS-MS using a minimum detection limit (MDL) of 10 parts per billion.

Sample Storage, Shipment, and Laboratory Analysis

All sample containers will be labeled for proper identification. Muscle, liver, and swab samples will be stored in a freezer immediately upon sampling and kept frozen until shipment to the laboratory. Samples will potentially be stored frozen up to 2 months prior to shipment. Thick-walled Styrofoam coolers and dry ice will be used to ship samples overnight to the Cornell Animal Health Diagnostic Center (AHDC).

Literature Cited

Miller, J. A. et al. 1983. Release of pyrethroids from insecticidal ear tags. Journal of Economic Entomology 76:1335-1340.