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THE IMPORTANCE OF AGRICULTURAL TIRE HABITATS FOR MOSQUITOES OF PUBLIC HEALTH IMPORTANCE IN NEW YORK STATE

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ABSTRACT. The presence of mosquito species breeding in agricultural "bunker tires" on dairy farms in New York state was evaluated. Twelve species of mosquitoes (*Aedes vexans, Anopheles barberi, An. punctipennis, An. perplexens, Culex pipiens pipiens, Cx. restuans, Cx. salinarius, Cx. territans, Toxorhynchites rutilus septentrionalis, Ochlerotatus atropalpus, Oc. japonicus japonicus,* and *Oc. triseriatus*) were collected from 8 New York state dairy farms in 2001 and from 17 dairy farms in 2002. All but 2 of these species have been found naturally infected with arboviruses and could be important vectors affecting human and animal health. Because of the potential public and animal health importance of the invasive species *Oc. j. japonicus,* active surveillance to identify breeding sites and distribution of this mosquito is essential. In 2001, *Oc. j. japonicus* was recovered from 5 of 8 central New York dairy farms surveyed. In 2002, *Oc. j. japonicus* was recovered from 4 of the same dairy farms plus an additional dairy out of 5 surveyed. This species appears to be established on dairy farms in the south, central, and eastern regions of New York state, with greatest abundance in the southeastern region. A single *Oc. j. japonicus* larva was collected from the northern region on the final sampling date in September 2002. Our data demonstrate that agricultural tire habitats can be productive breeding sites for arbovirus vectors. As a consequence, these habitats should not be ignored in vector control and surveillance programs.

KEY WORDS Dairy farms, public health, West Nile virus, mosquito vector, *Ochlerotatus j. japonicus,* invasive species

INTRODUCTION

The majority of large (200+ head) dairy farms in New York state use thousands of used tire casings to secure large tarps in place that cover bunker silage storage. This practice safeguards the anaerobic conditions needed for forage preservation (Mongeon 2000). However, the role that bunker tires play in production of mosquito vectors of West Nile virus (WNV) in New York state has never been examined. Many potential WNV vectors breed in tires and other types of man-made containers.

An additional risk has become apparent in recent years with the introduction of the exotic container-breeding mosquito *Ochlerotatus japonicus japonicus* (Theobald) into the United States and its subsequent spread through the eastern United States and the threat of *Aedes albopictus* (Skuse) expansion into the state. The 1st reported *Oc. j. japonicus* collections in the United States were recovered from New Jersey and New York in the late summer of 1998 (Peyton et al. 1999). Since its initial discovery, this invasive species has also been reported from several sites in New York (Falco et al. 2002, Oliver et al. 2003), Connecticut (Munstermann and Andreadis 1999), Maryland (Sardelis and Turell 2001), Ohio, Pennsylvania, and Virginia (Harrison et al. 2002, Sardelis et al. 2002a). It is possible that *Oc. j. japonicus* was present in some states as early as 1992 but was not detected (Andreadis et al. 2001). *Ochlerotatus j. japonicus* is reported to feed on birds and mammals, including humans (Kamimura 1968, Miyagi 1971, Tanaka et al. 1979). This mosquito has been shown to be a competent vector of Eastern equine encephalitis virus (Sardelis et al. 2002a), La Crosse virus (Sardelis et al. 2002b), and St. Louis encephalitis virus (Sardelis et al. 2003) and is a highly competent vector of WNV. In fact, Sardelis and Turell (2001) and Turell et al. (2001) demonstrated that *Oc. j. japonicus* is 2–4 times more competent as a vector of WNV than *Culex pipiens pipiens* (L.). It is likely that *Oc. j. japonicus* could play a role in arbovirus transmission in the northeastern United States; consequently, active surveillance to identify breeding sites and distribution of this species is essential. The potential of discarded tires and tire casings to harbor human disease vectors highlights the need to learn more about variation in species composition and ecology of mosquitoes breeding in these habitats. To address the importance of agricultural tire habitats as WNV vector breeding sites, a survey was conducted on New York state dairy farms over 2 years.

MATERIALS AND METHODS

Larval surveys: Surveys for mosquito larvae and pupae were conducted every 2 weeks from early June through late September 2001 and from late May through mid-October 2002. The tires sampled during both years of this study were piled or tossed adjacent to the bunker silage area, where the majority of tires on dairy farms are stored until reused when the bunkers are again filled during late summer and early fall (Fig. 1). Samples were collected from rimless used tire casings on 8 dairy farms in central New York in 2001 (Cayuga, Cortland, On-

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Fig. 1. Typical tire pile on a New York dairy farm.

ondaga, and Tompkins counties; Fig. 2). In 2002, mosquitoes were sampled from 3 dairy farms at each of 4 other regions across New York state. Areas sampled included farms in Wyoming County (western New York), St. Lawrence County (northern New York), Montgomery County (east-central New York), and Dutchess County (southeast New York). Additionally, 5 of the 8 central New York dairy farms sampled in 2001 were again sampled in 2002 (Fig. 2). Dairy farms in each region were a minimum of 10 km apart and had used tire piles as a result of the silage management practices discussed previously.

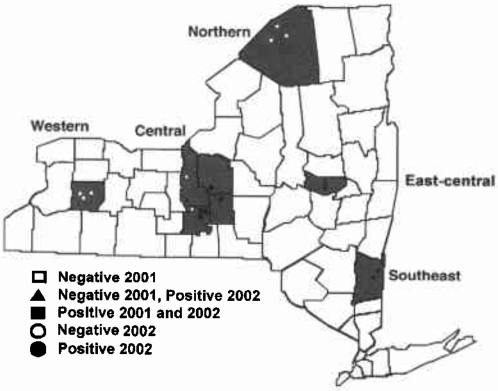

Fig. 2. Results of *Ochlerotatus j. japonicus* sampling from 8 counties in New York state, 2001 and 2002.

Mosquito larvae were sampled from tires with a white ladle (90 ml, 3 × 8 × 7 cm). Tires were sampled until collections were obtained from 10 different tires per farm. Because of personnel staffing and laboratory space challenges, following each Wyoming County collection, as well as September collections in Montgomery and St. Lawrence counties, mosquito larvae were removed and placed in vials of 70% ethanol. At other sites, mosquitoes were held for adult emergence in plastic cups (473 ml, Solo, Urbana, IL) that were fitted with emergence cones. Emerged mosquitoes were aspirated from containers daily and frozen at −20°C for future identification.

Adult surveys: Carbon dioxide–baited CDC miniature light traps (John W. Hock Company, Gainesville, FL, with the light bulb removed) were operated 1 night each week at the central New York sites during 2002. One trap was placed at each farm's tire pile and positioned 1.5 m above the

Table 1. Species and regional composition percentage of immature mosquitos collecting during 2001 and 2002 from tire casings on dairy farms in 5 areas of New York state.[1]

	Species composition (%)					
		2002				
	2001 Central	Central	Western	Northern	East-central	Southeastern
Aedes vexans	<1.0	—	—	—	—	<1.0
Anopheles barberi	—	—	<1.0	—	—	<1.0
An. perplexens	—	—	—	—	—	<1.0
An. punctipennis	<1.0	<1.0	<1.0	—	—	<1.0
Culex p. pipiens	4.6	9.3	7.6	36.3	25.4	11.3
Cx. restuans	35.6	51.2	91.8	50.1	55.7	33.9
Cx. salinarius	11.3	7.8	<1.0	4.1	16.0	5.0
Cx. territans	—	<1.0	<1.0	—	—	—
Cx. spp.	47.6	25.0	<1.0	9.0	<1.0	3.4
Ochlerotatus atropalpus	—	—	<1.0	<1.0	1.3	—
Oc. j. japonicus	<1.0	5.5	—	<1.0	1.2	42.2
Oc. triseriatus	<1.0	<1.0	—	—	—	3.3
Toxorhynchites rutilus septrionalis	—	—	—	—	—	<1.0
No. of specimens	10,597	8,991	13,559	1,974	2,946	2,865

[1] Central 2001 consisted of 8 dairy farms sampled on 9 dates; central 2002 consisted of 5 dairy farms sampled on 9 dates; northern 2002 and east-central 2002 each consisted of 3 dairy farms sampled on 9 dates; western 2002 consisted of 3 dairy farms sampled on 8 dates; southeastern 2002 consisted of 3 dairy farms sampled on 11 dates.

ground and baited with 0.5 kg of dry ice contained in a slow-release cooler. Traps were set up at ~1800 h (Eastern Daylight Savings Time [EDT]) and retrieved no later than 1000 h EDT the following day. Captured mosquitoes were placed in coolers containing wet ice, returned to the laboratory, frozen at −20°C, and identified to species. Because of limited resources, trapping at sites outside of central New York was conducted every 2nd week on the night prior to larval sampling. All adult Wyoming County and September collections from Montgomery County were frozen at −20°C and returned to Cornell University for identification. Adult and larval specimens were identified to species according to published keys (Means 1979, 1987; Darsie and Ward 1981; Darsie 2002).

RESULTS

Twelve species of mosquitoes were collected from tires on dairy farms, including *Ae. vexans* (Meigan), *Anopheles barberi* Coquillett, *An. perplexens* Ludlow, *An. punctipennis* (Say), *Cx. p. pipiens, Cx. restuans* Theobald, *Cx. salinarius* (Coquillett), *Cx. territans* Walker, *Toxorhynchites rutilus septentrionalis* (Dyar and Knab), *Oc. atropalpus* (Coquillett), *Oc. j. japonicus,* and *Oc. triseriatus* (Say; Table 1). The dominant tire-breeding species collected on dairy farms in our study was *Cx. restuans* in all regions, with the exception of the southeastern region. In the western region, >91% of specimens collected were *Cx. restuans.* The 2nd most commonly collected species was *Cx. p. pipiens.* In the southeastern region, *Oc. j. japonicus* was the most commonly collected species, accounting for >42% of reared adults.

In 2001, *Oc. j. japonicus* was recovered only from the 5 easternmost farms in the central region (Cortland, Onondaga, and Tompkins counties). Specimens of *Oc. j. japonicus* were recovered monthly from June to September. The greatest abundance of *Oc. j. japonicus* in collections was in August (3.4 specimens per farm per collection). Between 2001 and 2002, the percentage of *Oc. j. japonicus* recovered at farms positive in 2001 increased from 1.6 to 7.5% of all larvae recovered.

During the 2002 survey, we collected and reared to the adult stage a total of 23 immature *Oc. j. japonicus* from a Cayuga County dairy farm (central region), where the species was not detected in 2001. No specimens of *Oc. j. japonicus* were found in tire casings on dairy farms in the western region of New York, whereas a single larva was collected from a farm in the northern region on the final sampling date (Fig. 1). Overall, the greatest numbers of *Oc. j. japonicus* per farm per sampling date were recovered from farms in the southeastern region (36.7) followed by farms in the central region (10.9), with only 1.3 specimens per farm per date collected from farms in the east-central region. Recovery of immature *Oc. j. japonicus* occurred in the southeastern region in each month sampled and peaked in September when 76 specimens per farm were collected, whereas abundance in the central and east-central regions peaked in August (Fig. 3).

The monthly composition of the 4 primary mosquito species collected is presented in Fig. 4. The relative abundance of *Cx. restuans* fell from 70% of larvae collected in May and June to approximately 50% in October. The recovery of *Cx. p. pipiens* increased from 13% in May to 35% in October of total collections. The prevalence of *Oc. j.*

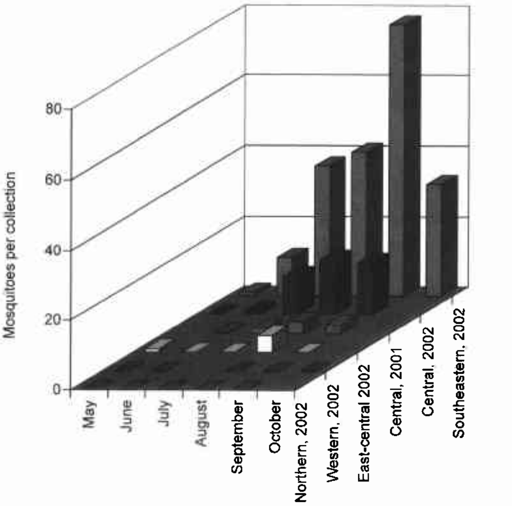

Fig. 3. Mean number of *Ochlerotatus j. japonicus* collected per farm visit from larval samples of used tire casings at dairy farms in 1 region of New York state in 2001 and 5 regions in 2002. Missing data bars represent no collections because of study length or drought.

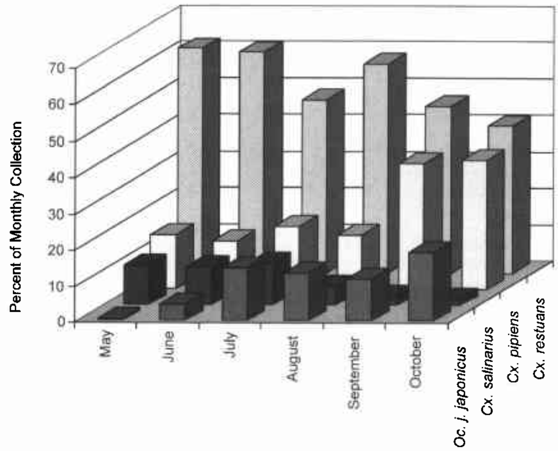

Fig. 4. Percent monthly composition of primary mosquito species collected as larvae from used tire casings at dairy farms in 5 regions of New York state in 2002 (minor and generic-only collections not shown).

japonicus increased from <1% in May to >19% in October.

Approximately 7 adult mosquitoes per trap night per farm were recovered at dairy farms in the western, east-central, and southeastern regions from the CO_2-baited CDC miniature light traps (Table 2). In traps with fans operating throughout the night at central region dairy farms, nearly 11.3 adult mosquitoes per trap night per farm were recovered. For reasons that are not clear, only 2 adult specimens were collected on northern region dairy farms during the entire survey period.

With the exception of southeastern region farms, at which *Coquillettidia perturbans* Walker was most often collected, *Cx. p. pipiens* and *Cx. restuans* were the predominant adult mosquito species collected in the western, central, and east-central regions (Table 2). Few *Aedes, Anopheles,* and *Ochlerotatus* specimens were collected in any region. In total, 12 mosquito species were collected in CO_2-baited CDC miniature traps.

Few adult *Oc. j. japonicus* were recovered from CO_2-baited CDC miniature traps, even in areas where larval abundance was high. During 240 trap hours (TH), no *Oc. j. japonicus* was collected in the northern region, the east-central region (288 TH), or western region (308 TH). In the southeastern region (336 TH) and central region (950 TH),

Table 2. Number of adult mosquitos collected during 2002 from CO_2-baited light traps placed near bunker tire piles on dairy farms in 5 regions of New York state.[1]

	No. of adults collected				
	Central[2]	Western	Northern	East-central	Southeastern
Aedes vexans	1	0	0	3	0
Anopheles barberi	2	0	0	0	3
An. punctipennis	1	3	0	0	0
An. quadrimaculatus	12	0	0	0	0
Coquillettidia perturbans	37	11	0	0	134
Culex p. pipiens/restuans	187	90	1	100	4
Cx. salinarius	12	1	0	22	2
Cx. spp.	16	48	—	—	—
Ochlerotatus aurifer	0	0	1	0	1
Oc. fitchi	0	1	0	0	0
Oc. j. japonicus	1	0	0	0	3
Oc. triseriatus	0	0	0	0	6
Ae./Oc. spp.	1	1	—	—	—
No. of specimens	270	155	2	125	153

[1] Central consisted of 13 trap dates on 5 dairy farms; western and southeastern each consisted 7 bimonthly trap dates on 3 dairy farms; east-central and northern consisted 6 and 9 bimonthly trap dates each on 3 dairy farms, respectively.

[2] Malfunctions resulted in successful trap operations from 24 of 65 trap nights.

3 and 1 adult *Oc. j. japonicus,* respectively, were collected in CDC miniature light traps.

DISCUSSION

New York is the 3rd largest dairy producing state, with over 6,680 dairy farms, many of which use economical and large-capacity bunker silos to store forage (Anonymous 2004, Lanyon et al. 2004). As dairy farms continue to get larger, bunker use will likely increase (Lanyon et al. 2004). Most farms with >200 cows currently use bunker silos because of convenience and cost, and nearly all of these farms use tire casings to hold the plastic covers in place.

The amount of water observed in tires varied throughout the summer. Therefore, during dry spells, larvae and pupae were generally not present for lack of water. However, immature mosquitoes were readily collected from these same tire casings shortly following rainfall events. Compared with other mosquito species collected, *Oc. j. japonicus* tended to be recovered from discolored water, which might be indicative of higher organic content, as well as from tires with diminishing water levels.

All but 2 of the species of mosquitoes recovered from bunker tires on dairy farms in New York state are considered of medical or veterinary importance. *Aedes vexans* is considered a vector of eastern equine encephalomyelitis virus (EEE) (Scott and Weaver 1989) and has been found naturally infected with WNV (CDC 2000, White et al. 2001). *Anopheles barberi* has been found infected with WNV as has *An. punctipennis* (CDC 2003). *Culex p. pipiens* is considered the most important enzootic vector of WNV (CDC 2000, White et al. 2001). *Culex restuans* and *Cx. salinarius* are also potential bridge vectors of WNV (Turell et al. 2001). *Culex salinarius* has also been implicated as a bridge vector of EEE (Vaidyanathan et al. 1997). *Culex territans* has been found naturally infected with WNV in New York state (Kulasakera et al. 2001, CDC 2003). In addition, *Oc. atropalpus, Oc. j. japonicus,* and *Oc. triseriatus* have all been found naturally infected with WNV (CDC 2000, Kulasakera et al. 2001, White et al. 2001, CDC 2003).

The predominant mosquito larval species collected from tires on southeastern dairy farms was *Oc. j. japonicus,* which accounted for 42% of recovered specimens. This species appears to be well established in that region and might be competitively excluding local tire-breeding species, such as *Cx. p. pipiens* and *Cx. restuans.* The number of *Oc. j. japonicus* collected from tires increased from 2001 to 2002 on central region dairy farms (Fig. 2). Additionally, the appearance of *Oc. j. japonicus* on a Cayuga County farm in 2002 marked the westernmost recovery of this species on a dairy farm in New York state. Specimens of *Oc. j. japonicus* were not found among >13,000 mosquito larvae recovered and identified from western region dairy farms.

CDC miniature traps were not useful for adult collection of *Oc. j. japonicus.* Greater recovery of adult *Oc. j. japonicus* from gravid traps versus CO_2-baited traps has been described recently by Scott et al. (2001), who collected as many as 6.25 *Oc. j. japonicus* per gravid trap night. Falco et al. (2002) reported that *Oc. j. japonicus* made up only 1.5% of specimens in light traps while accounting for 18.1% of species in gravid traps. CDC light trap–based surveillance programs that want to detect *Oc. j. japonicus* will need to incorporate other collection methods, such as the gravid trap.

Ochlerotatus j. japonicus is moving rapidly across New York state and into other areas of the United States (Oliver et al. 2003). Given the demonstrated vector competence of this species for several human pathogens (Sardelis and Turell 2001; Turell et al. 2001; Sardelis et al. 2002a, 2002b, 2003) and the potential of this species to feed on human and other animal blood (Kamimura 1968, Miyagi 1971, Tanaka et al. 1979, Peyton et al. 1999), further research is needed to determine the potential of *Oc. j. japonicus* to competitively displace native species breeding in container habitats such as tires.

Dairy producers need to be made aware of the mosquito-breeding risk that water-containing tire casings pose, especially for those held haphazardly in piles. Suggestions for alternatives to the use of uncut tire casings are available (Lanyon et al. 2004). These include using sidewall disks of bias-ply truck tires and heavy equipment tire beads. The use of alternative storage procedures, such as holding tires under tarps, are rarely practiced but could dramatically reduce mosquito production.

Our results document that several mosquito species of public and animal health importance are breeding in agricultural habitats such as tire casings on New York state dairy farms. Furthermore, our research supports the inclusion of dairy farms in public health surveillance programs for rural areas. Finally, dairy producers need to identify alternatives to standard tire casings as a component of their feed storage program and properly dispose of existing tires that serve to support mosquito breeding.

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