THE MOSQUITOES ATTRACTED TO TURTLES [1]

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INTRODUCTION. A knowledge of the mosquito species that commonly feed on reptilian hosts is necessary to better understand enzootic virus cycles in nature. Reptiles have long been considered possible reservoirs of arboviruses, and numerous reptilian recoveries of both eastern and western encephalitis viruses have been reported. Naturally infected turtles have repeatedly been found in New Jersey. Goldfield and Sussman (1967) reported the recovery of EE virus from the brain of a snapping turtle and from the blood of one painted turtle and two box turtles WE virus was isolated from the blood o a northern diamondbacked terrapin anc a box turtle. In the same studies, neutral ization antibodies to EE were found ir 48 of 1,460 turtles tested and neutraliza tion antibodies to WE were found in 4(of 1,447 turtles tested.

The number of mosquito species on the eastern seaboard that do feed on turtle: is not presently known. Bait studies have not suggested that turtle feeding is com mon for any mosquito other than *Culex territans*. Hayes (1961) found only limited turtle feeding by *Culex pipiens Culex restuans* and *Culex salinarius* ir Massachusetts. Several mosquito species were attracted to turtles in his studies bu did not engorge. *Aedes canadensis* dic

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not readily come to turtle hosts but showed a high rate of engorgement once attracted. Murphey *et al.* (1967) reported limited feeding on turtles by *Aedes sollicitans* and *Anopheles quadrimaculatus* in bait studies conducted in Delaware. *Culex territans,* a species known to feed primarily on cold-blooded animals, was the only mosquito which readily accepted turtles in the Delaware investigations.

Mosquitoes have been noted feeding on turtles under natural conditions on several occasions. Hayes (1965) observed approximately 50 *Aedes canadensis* hovering and engorging upon a box turtle in Illinois. Nolan *et al.* (1965) aspirated 37 *Aedes canadensis* and a single *Aedes triseriatus* from a painted turtle and 6 *Aedes canadensis* from a box turtle in Maryland. Two of the mosquitoes thus obtained were distended with freshly ingested blood and were assumed to have fed on the turtle. DeFoliart (1967) netted 19 mosquitoes from a Blanding's turtle in Wisconsin. Seventeen of these were *Aedes canadensis,* 6 of which were engorged. One *Aedes cinereus* and one member of the *Aedes communis* group were also taken but were presumed to have been attracted to the vicinity by the collectors' presence.

The writers have repeatedly observed mosquitoes attacking turtles in New Jersey. In all cases, *Aedes canadensis* has been the species most encountered. A compilation of these observations and subsequent experimentation are the subjects of this paper.

MATERIALS AND METHODS. Mosquito collections were taken both from turtles that were encountered in the wild and from turtles that were deliberately exposed. All turtles encountered in the field over a 6-year period in southern New Jersey were routinely examined for mosquitoes. Collections were made by placing a standard insect net over the reptile and trapping all mosquitoes that were hovering about the animal. Several quick sweeps of the net concentrated the mosquitoes which were then killed in a cyanide jar and placed in vials with information concerning date, location and turtle species. Mosquito identification was later performed at the labortory where engorged specimens were subjected to serological tests to determine unequivocally the source of host blood.

For field exposures, ten eastern box turtles, *Terrapene carolina carolina,* were captured and held as test animals. In early trials, turtles were exposed within circular wire enclosures approximately 2 ft. in diameter. Later each specimen was fitted with a wire ring to which a leash could be attached. Rings, made from small diameter wire, were looped through a small hole drilled in the edge of the carapace near the hindquarters of the animal. Each turtle was then tethered on a 3-foot length of nylon fishing cord fitted with a snap swivel that could be clipped to the wire ring on the turtle's shell. Using this method, turtles were free to walk within a 6-foot diameter circle. In most cases, turtles were active, continually walking in a circle which soon became a well demarcated path. Late in the afternoon and when mosquito activity was high during the day, turtles were frequently found burrowed into the leaf litter. Mosquito collections from tethered turtles were made, using an insect net as previously described.

Tethered box turtles were stationed daily at each of five different locations near Woodbine, New Jersey, during the fall of 1967. Two of the stations were in dried depressions within climax woodlands of white oak. These dried depressions were woodland pools which were assumed to have produced *Aedes canadensis* earlier in the season. Other locations included a red maple woodland, a white cedar swamp and a white pine plantation. Because locations were separated by several miles, observations were not performed at standard time intervals. Collections were usually begun in the daytime, approximately 2 hours after the turtles had been exposed, but several collections were also made after dark from turtles which had been exposed up to 6 hours.

Mosquito populations in the Woodbine area were concurrently sampled with resting boxes and light traps. Collections from 50 resting boxes, distributed throughout the area, were taken each morning that turtles were exposed. Five standard and five battery operated New Jersey light traps were activated at dusk following turtle exposure.

RESULTS AND DISCUSSION: (1) Collections From Turtles Encountered in the Wild. The results of 17 mosquito collections from 5 different turtle species are listed in Table 1. Each collection represents one isolated instance where a turtle in the wild was found being fed upon by mosquitoes. *Aedes canadensis*, a common spring mosquito, was the only species collected from turtles early in the season. No mosquitoes were detected feeding on turtles during the mid-summer months of any year. Excessive rains late in the summer of 1967 triggered a late season brood of *Aedes canadensis*, and nearly every turtle encountered after this brood emerged was being attacked by mosquitoes. Collections at this time revealed that *Aedes canadensis* was the most common mosquito feeding on turtles, but small numbers of five other species were collected as well. *Aedes atlanticus*, a rare species in New Jersey, was taken on three occasions. A single engorged specimen was subjected to serological tests and was shown to have fed on a turtle host. A description of the *Aedes atlanticus* collections has been published elsewhere (Crans, 1968). *Aedes triseriatus* was taken on two occasions, but

TABLE 1.—Mosquitoes collected from turtles over a six-year period.

Turtle host	Date	Mosquito species	Engorged	Unengorged	Total
		Early Season Collections			
Snapping turtle[1]	6/62	*Aedes canadensis*	4	0	4
Redbellied turtle[2]	6/62	*Aedes canadensis*	0	6	6
Eastern box turtle[3]	6/63	*Aedes canadensis*	15	4	19
Eastern box turtle	6/63	*Aedes canadensis*	20	10	30
Eastern box turtle	6/66	*Aedes canadensis*	10	2	12
Spotted turtle[4]	6/67	*Aedes canadensis*	4	3	7
		Late Season Collections			
Spotted turtle	8/67	*Aedes canadensis*	0	79	79
		Aedes atlanticus	0	3	3
Eastern mud turtle[5]	8/67	*Aedes canadensis*	5	11	16
Eastern box turtle	8/67	*Aedes canadensis*	0	2	2
		Aedes sollicitans	0	4	4
		Culex sp. ♂	..	1	1
Eastern box turtle	8/67	*Aedes canadensis*	0	12	12
Eastern box turtle	8/67	*Aedes canadensis*	22	165	187
		Aedes atlanticus	1	2	3
Eastern box turtle	8/67	*Aedes canadensis*	9	21	30
		Aedes sollicitans	0	1	1
Eastern box turtle	8/67	*Aedes canadensis*	2	10	12
		Aedes triseriatus	0	1	1
Eastern box turtle	8/67	*Aedes canadensis*	23	103	126
		Aedes atlanticus	0	1	1
Eastern box turtle	8/67	*Aedes canadensis*	11	68	79
		Aedes cantator	0	1	1
		Aedes triseriatus	0	1	1
Eastern box turtle	8/67	*Aedes canadensis*	4	9	13
		Culex salinarius	0	1	1
Eastern box turtle	9/67	*Aedes canadensis*	4	5	9

[1] *Chelydra serpentina.*
[2] *Pseudemys rubriventris.*
[3] *Terrapene carolina carolina.*
[4] *Clemmys guttata.*
[5] *Kinosternon subrubrum subrubrum.*

neither specimen had taken a blood-meal. *Aedes sollicitans, Aedes cantator,* and *Culex salinarius* were taken in small numbers, but no engorged specimens were included. *Aedes sollicitans* were particularly numerous when late season collections were being made, and it is possible that the few specimens taken were attracted to the collectors rather than to the turtles. The collection technique involved sweeping the insect net several times to concentrate the mosquitoes in the net bag, and although efforts were made to exclude airborne mosquitoes, specimens could have been swept into the collection.

The majority of turtles sampled were encountered on roads or basking in rain-pools. *Aedes canadensis* were observed to hover about a turtle, usually in a dense cloud which was visible from a distance of many yards. Unless a net was quickly placed over the turtle, mosquitoes would attack the collectors in a feeding frenzy never before encountered by these authors with *Aedes canadensis.* Several specimens with a partial blood-meal were observed continuing the meal on the collectors, indicating that the species would accept multiple hosts. Observation revealed that although some mosquitoes probed between the scutes of a turtle's carapace, most feeding occurred about the head, neck and legs. When turtles withdrew their head and legs, mosquitoes were often crushed. Some mosquitoes were trapped under the carapace of box turtles when the hinged plastron was closed. After collections were made, turtles would cautiously extend their heads, revealing numerous mosquitoes trapped in the folds of skin on the neck. Often, mosquitoes would fly out from under the carapace when a turtle began to move. Mosquitoes which were crushed or trapped under turtles' shells, as well as those which left the turtles to feed on the collectors, were not included in Table 1.

(2) Collections from Tethered Box Turtles. The mosquitoes collected from box turtles tethered at five different locations near Woodbine, New Jersey, are listed in Table 2. In all collections and in all areas, *Aedes canadensis* was the species taken most often on turtles, comprising more than 99 percent of the total. The exposures made in dried woodland depressions thought to have produced *Aedes canadensis* earlier in the season (stations #1 and #4), yielded the greatest numbers of mosquitoes. Stations #1 and #4 averaged 70 and 40 mosquitoes per collection respectively, even though exposures at these two

TABLE 2.—Mosquitoes collected from tethered box turtles at five different New Jersey habitats.

Mosquito species	Habitat [1] Station #1 Oak Woods (24)	Station #2 Maple Woods (11)	Station #3 Cedar Swamp (7)	Station #4 Oak Woods (24)	Station #5 Pine Plantation (6)	Total all areas (72)
Aedes canadensis	1715/430[2]	235/76	26/15	1006/423	36/21	3018/965
Aedes sollicitans	4/0		1/0	9/1**		14/1**
Aedes triseriatus	4/0					4/0
Aedes vexans				2/0		2/0
Aedes cantator	1/0	1/0				2/0
Coquillettidia perturbans			2/1*			2/1*
Culiseta melanura	1/1*				2/1*	3/2*
Culex restuans				1		1
Culex sp. ♂						

[1] Number under each station indicates the number of collections performed.
[2] Number attracted/number engorged.
* Contained avian blood.
** Contained human blood.

areas continued late into the season when mosquito populations were low. At these two collection sites, over 100 *Aedes canadensis* were collected from a single turtle on each of nine different occasions. The highest number of mosquitoes taken in a single collection occurred at Station #1 when 569 *Aedes canadensis* and a single *Aedes triseriatus* were netted at one time from one turtle. Turtles stationed in the cedar swamp attracted the fewest mosquitoes, averaging slightly over 4 mosquitoes per collection.

Data collected from turtles suggested that *Aedes canadensis* might be the most numerous mosquito at the collection sites, but light trap and resting box captures revealed that this species actually made up only a small percentage of the total population (Table 3). Even though five other mosquito species were collected in greater numbers in the Woodbine area, *Aedes canadensis* was the only mosquito which actually fed on the reptiles. In all probability, some of the specimens contained the blood of other animals, but random serological tests on over 100 individuals failed to reveal other than turtle feedings. The numbers of engorged *Aedes canadensis* taken during these studies prohibited the serological testing of all specimens.

Freshly ingested blood was detected in several other mosquito species (Table 2), but serological tests revealed that in all cases, the blood-meals had been taken from other animals. The two *Culex restuans* and the single engorged *Culiseta melanura* that were taken from turtles contained avian blood-meals. The only engorged *Aedes sollicitans* taken from a turtle had fed on a human.

These findings cast doubt on the validity of the bait trap technique when freshly ingested blood is used as the only criterion for engorgement on the test animal. Data in Table 2 suggest that some mosquitoes may be attracted to the host animal even after they have fed to repletion on a different host. In addition to the attractiveness of the test animal, most bait trap designs also provide mosquitoes with the shelter of a resting container, and it is likely that this combination of factors might attract fully engorged mosquitoes. An unbaited trap used as a control does not control both of these factors. Engorged specimens which had fed on other hosts were collected in the present studies even though the turtles were exposed without the confinement of a structure.

It is possible that some feedings attributed to a particular test animal in bait

TABLE 3.—Mosquito fauna in the Woodbine area as indicated by resting box and light trap collections.

Mosquito species	Number mosquitoes collected			Percent of total catch
	Resting box[1]	Light trap[2]	Total	
Culiseta melanura	2180	1735	3915	36.2
Culex salinarius	66	2096	2162	20.0
Anopheles bradleyi (complex)	162	1137	1299	12.0
Aedes sollicitans	2	1154	1156	10.7
Culex territans	441	39	480	4.4
Aedes canadensis	11	451	462	4.3
Culex pipiens	389	14	403	3.7
Aedes cantator	2	352	354	3.3
Culex restuans	100	154	254	2.4
Anopheles punctipennis	181	4	185	1.7
Aedes vexans	0	117	117	1.1
Anopheles quadrimaculatus	19	0	19	1.0
Other *	5	7	12	1.0
Total	3558	7260	10,818	100.0

[1] 50 Resting boxes.

[2] 5 Standard and 5 battery operated New Jersey Light Traps.

* *Culiseta minnesotae, Culiseta inornata, Uranotaenia sapphirina, Aedes triseriatus, Aedes taeniorhynchus, Mansonia perturbans, Psorophora ferox* and *Anopheles barberi.*

trap exposures are actually derived from other sources, particularly in those cases where small numbers of mosquitoes are attracted to "unusual" hosts. New host records based on bait trap results without observational or serological confirmation may not always be indicative of a mosquito's true feeding habits.

The high numbers of *Aedes canadensis* that were collected from turtles suggested that the reptiles provided some form of attractant which might have been selective for this mosquito. During these investigations, further evidence of a scent or residue which may have served as the attractant was observed. On many occasions, mosquitoes were observed gathering at the collection site after the turtles had been removed. In addition, on three separate occasions, a turtle had escaped before collections could be made, and in each case mosquitoes were observed hovering about the abandoned depression in the leaf litter where the turtle had previously rested. Some mosquitoes were resting in the leaf litter, but in all cases the majority of specimens were actively hovering over the spot just as they did when a turtle was present.

Table 4 lists the mosquitoes which were captured from abandoned depressions in leaf litter after turtles had escaped. As many as 133 specimens were captured in this manner, and *Aedes canadensis* was the only mosquito species represented. The fact that engorged specimens were taken in this manner indicated that some mosquitoes remained at the site of feeding, but the high incidence of unengorged mosquitoes which hovered about the abandoned depression suggested that some form of residue from the turtle was attractive to *Aedes canadensis,* and possibly only to *Aedes canadensis.* Studies are presently being planned to test these hypotheses fully.

TABLE 4.—Numbers of *Aedes canadensis* attracted to abandoned depressions formed by box turtles in leaf litter.

Observation no.	Engorged	Unengorged	Total
1	16	61	77
2	7	115	122
3	19	114	133

SUMMARY AND CONCLUSION. Mosquitoes were collected both from turtles that were encountered in the wild and from turtles that were deliberately exposed at five different habitat locations. *Aedes canadensis* dominated all mosquito collections from turtles, even though light trap and resting box records showed that several mosquito species were more numerous at each location. *Aedes canadensis* and *Aedes atlanticus* were the only species found to feed on turtles during the investigations. Several other mosquito species with freshly ingested blood were collected from turtles, but serological tests revealed that in all cases the blood-meals had been derived from other animal sources. The attraction of *Aedes canadensis* to abandoned depressions in leaf litter formed by turtles indicated that the reptiles left some form of residue which was attractive to the mosquitoes.

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