

a total of 78 lots of *Dermanyssus* and *Liponyssus* mites from wild birds' nests collected in Weld County, May through August, 1950. The bird nest sources of the mites and the species tested are presented in Table I.

The tests were made on approximately 500 mites sealed for each nest, except where this number was not available, as at the first part of the nesting season. Mite species determinations were based on samples of usually 10 to 30 specimens preserved in alcohol from each nest. The methods of collecting and of testing as described above in the recovery of WEE virus were used in all tests.

Identification of the mites from the virus-infected lot was made on 34 specimens, all *D. americanus*, 6 males, 26 females and 2 nymphs. This is the first definite recovery of a neurotropic virus from *D. americanus*, although Reeves and his associates(1) obtained evi-

dence of a possible infection of this mite with WEE virus in Kern County, California, where, in recoveries of the virus from *Liponyssus sylviarum*, they found *D. americanus* included with *L. sylviarum* in one pool of mites found infected with WEE virus. Natural infections with this virus have been reported from the chicken mite *Dermanyssus gallinae* by Sulkin(2) and from the tropical fowl mite *Liponyssus bursa* by Sulkin and Izumi(3).

Summary. Virus of Western equine encephalomyelitis was isolated from mites (*Dermanyssus americanus*) collected in Weld County, Colo. in July 1950.

Grateful acknowledgment is given to Assistant Sanitarian (R) John E. Lane of the Entomological Section, Laboratory Services, CDC, Atlanta, Ga. for identification of the mites.

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3. Sulkin, S. E., and Izumi, E. M., *Proc. Soc. Exp. Biol. and Med.*, 1947, v66, 249.

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Recovery of Virus of Eastern Equine Encephalomyelitis from a Mosquito, *Culiseta melanura* (Coquillett)* (18790)

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Although the virus of eastern equine encephalomyelitis has been transmitted by several species of *Aedes* mosquitoes in the laboratory(1,2) and is generally considered to be arthropod-borne, isolations from arthropods in nature have been few. Howitt *et al.*(3) made the first isolation of EEE virus from wild-caught arthropods when they recovered

it from common chicken mites (*Dermanyssus gallinae* (De Geer)) and chicken lice (*Eomenacanthus stramineus* (Nitzsch)), collected in Tennessee in 1947. The only other isolation of EEE virus from arthropods in nature has been from a mosquito, *Mansonia perturbans* Walker in Georgia, also reported by Howitt *et al.*(4).

Western equine encephalomyelitis, on the other hand, has been recovered from wild-caught *Culex tarsalis* Coquillett(5), *Culiseta inornata* (Williston), *Culex pipiens* Linnaeus and *Anopheles maculipennis freeborni* Aitken

* This note constitutes part of an investigation on encephalitis in Louisiana conducted by the Virus and Rickettsia Section, Montgomery, Ala.

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2. Davis, W. A., *Am. J. Hyg.*, 1940, v32, 45.

3. Howitt, B. F., Dodge, H. R., Bishop, L. K., and Gorrie, R. H., *Proc. Soc. Exp. Biol. and Med.*, 1948, v68, 622.

4. Howitt, B. F., Dodge, H. R., Bishop, L. K., and Gorrie, R. H., *Science*, 1949, v110, 141.

5. Hammon, W. McD., Reeves, W. C., Brookman, B., and Izumi, E. M., *J. Infect. Dis.*, 1942, v70, 263.

(6), *Aedes dorsalis* (Meigen) and *Culex stigmatosoma* Dyar(7), *Triatoma sanguisuga* Le Conte(8), *D. gallinae* (De Geer)(9), *Liponyssus bursa* (Berlese)(10), and *Liponyssus sylvium* (Canestrini and Fanzago)(11).

This note reports the isolation of EEE virus from another species of mosquito, *Culiseta melanura* (Coquillett). The infected lot consisted of two specimens collected by light trap in the Manchac Swamp near Ponchatoula, Louisiana, on August 17, 1950. It is of interest that these specimens were taken within a few hundred yards of the location where a purple grackle with EEE virus in its blood was shot June 19, 1950(12).

The specimens were removed alive from the light trap, sealed in a tube, and kept frozen on dry ice until processed. At the laboratory in Montgomery, Ala., they were identified, triturated in 1 ml of tryptose phosphate broth, and spun 20 minutes in the cold at 4000 RPM in an angle-head centrifuge. After adding 2 mg of streptomycin and 500 units of penicillin, a portion of the supernate was used to inoculate six 3-week-old Swiss mice intracerebrally. Neurological symptoms were observed in the mice late the second day, followed by death on the third day. No bacterial growth was observed in broth or on blood agar plates following inoculation with suspension of brain material from the dead mice.

A second intracerebral passage caused death of mice in 2-3 days. Brains from this passage had an LD₅₀ titer of 10⁻⁸, as calculated by the method of Reed and Muench, upon intra-

cerebral inoculation into mice(13). The infectious agent passed a Seitz EK filter.

Serum from a rabbit immune to the Decuir strain of EEE neutralized 3000 mouse LD₅₀'s of the new agent when serial 10-fold dilutions of infected brain were mixed with equal quantities of undiluted serum, incubated 18 hours at 4°C and inoculated intracerebrally into 3-week-old mice. WEE immune serum failed to have a neutralizing effect in tests conducted simultaneously. Sera from rabbits immunized against the new agent had a neutralization index of 1000 against the Decuir strain of EEE.

Guinea pigs immune to the Decuir strain of EEE resisted an intracerebral challenge of 10⁷ mouse LD₅₀'s of the new agent. Those immunized against the new agent resisted a similar intracerebral challenge of the Decuir strain. Similarly immunized animals succumbed to 10⁵ LD₅₀'s of WEE virus given intracerebrally.

Host range studies conducted in 10-day-old and adult mice, guinea pigs, hamsters, rabbits, and chickens revealed clinical courses similar to those of the equine encephalitides in these species. A pathological picture consistent with that produced by the arthropod-borne encephalitis viruses was found upon microscopic examination of brain sections of infected mice, guinea pigs, hamsters, and rabbits.

Confirmatory isolation from the original mosquito suspension was obtained by its re-inoculation into a second group of mice.

As very little is known of the adult habits of *C. melanura* and only a single isolation has been made, the possible significance of this species in the epidemiology of EEE cannot be evaluated at the present time.

Summary. The virus of eastern equine encephalomyelitis was isolated from mosquitoes (*Culiseta melanura* (Coquillett)) collected by light trap in Louisiana, August, 1950.

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