SURVEILLANCE OF ARBOVIRUS ACTIVITY IN IOWA, 1977.[1]

D. C. DORSEY,[2] W. A. ROWLEY,[3] Y. W. WONG,[2] J. P. BRINKER,[2] R. W. CURRIER[4] AND W. J. HAUSLER, JR.[2]

ABSTRACT. The State of Iowa maintains a continuing surveillance program for the detection of arbovirus activity. Results of laboratory studies of avian, human, and mosquito specimens collected during the summer of 1977 are presented. Cache Valley, Flanders, trivittatus, Turlock, and western equine encephalomyelitis arboviruses were isolated from 73 of the 613 pools of mosquitoes examined. Avian blood specimens collected from 41 species were examined for St. Louis encephalitis (SLE) and western equine encephalomyelitis (WEE) hemagglutination-inhibition (HI) antibodies. Both SLE and WEE antibodies were detected among the 1,037 avian sera tested. One case of WEE and three cases of California encephalitis in humans were serologically confirmed and are discussed. A survey to determine the incidence of encephalitis in equines revealed arbovirus activity throughout the state.

The State Hygienic Laboratory has monitored arbovirus activity in Iowa since the inauguration of an encephalitis surveillance program in 1965. That year, 6 cases of LaCrosse (California) encephalitis (LAC) were detected in children with central nervous system disease (Wong et al. 1970). Results of subsequent studies involving the collection of mosquitoes for virus isolation and the serologic testing of indicator animals placed in areas of known virus activity to monitor arboviruses in Iowa have been reported (Wong et al. 1971, 1973).

Since 1971, the surveillance program has been a cooperative effort with the Entomology Department of Iowa State University. Field and laboratory studies have been expanded to include collection sites throughout Iowa. In 1971, Cache Valley (CV), Flanders (FL), LaCrosse (LAC), St. Louis encephalitis (SLE), and western equine encephalomyelitis (WEE) viruses were isolated from 41 of 1,542 pools totaling 69,464 mosquitoes (Rowley et al. 1973). More recently, this program has been a combined effort of Iowa State University, the State Health Department, and the State Hygienic Laboratory. The isolation of arboviruses and detection of human cases of mosquito-borne encephalitis continue to be reported annually in Iowa (Wong et al. 1978).

A major outbreak of St. Louis encephalitis in the upper Mississippi Valley in 1975 focused the attention of local health officials on arbovirus activity in their communities. Programs to monitor vector populations for both mosquito control and virus activity have received renewed interest and concern by public health personnel. Because birds are the primary vertebrate hosts of *Culex* mosquitoes, the data reported in our surveillance program include the serologic survey of wild birds. Antibody detected in juvenile birds is considered indicative of recent viral transmission.

This report summarizes the results of studies made of avian sera, equine and human cases, and mosquito specimens collected for both virus isolation and mosquito control during the summer of 1977.

MATERIALS AND METHODS

Adult mosquitoes were collected for virus isolation concurrently in areas

[1] Journal Paper No. J-9155 of the Iowa Agriculture and Home Economics Experiment Station, Ames, Iowa, Project No. 2053.

[2] State Hygienic Laboratory, University of Iowa, Iowa City, Iowa 52242.

[3] Department of Entomology, Iowa State University, Ames, Iowa 50011.

[4] Iowa State Health Department, Lucas Office Building, Des Moines, Iowa 50319.

where ongoing mosquito population surveys were being conducted for mosquito control. Additional collection sites were selected in communities where arbovirus activity occurred. Mosquitoes were collected for virus isolation studies with CDC light traps baited with dry ice and operated by flashlight batteries. Mosquitoes were processed according to standard techniques (Sudia & Chamberlain 1967). Isolation of viruses in 24 to 48-hr-old suckling mice has been described (Wong et al. 1971). Standard New Jersey light traps equipped with a 40-watt bulb were used for mosquitoes population studies.

Human cases were verified as a routine procedure in the Virology Division of the State Hygienic Laboratory by examination of sera from suspected cases of central nervous system disease for complement-fixing (CF), hemagglutination-inhibition (HI), and (or) neutralizing (N) antibodies against LAC, SLE, and WEE antigens (Lennette & Schmidt 1969, U.S. Public Health Service 1965). Sucrose acetone extracted infected mouse brain antigens were used in both the CF and HI tests. The microtiter system was employed for the CF, HI, and N tests. Neutralization tests were performed in disposable microtiter plates, using baby hamster kidney (BHK-21) cells (Lennette & Schmidt 1969).

Birds were trapped with nylon mist nets and, occasionally, in baited cages. Birds were identified to species, and their ages (adult or juvenile) and other pertinent information were recorded. Approximately 0.2 ml of avian blood was collected from the jugular vein with a 26-gauge needle affixed to a 1-ml disposable syringe. The blood was added to 0.8 ml of field diluent (1% bovine albumin in phosphate buffered saline, pH 7.6). Sera were treated with protamine sulfate to remove acetone-insoluble inhibitors and then acetone extracted before testing for SLE and WEE hemagglutination-inhibition antibodies (Holden et al. 1966).

Data on cases of equine viral encephalitis were obtained from a retrospective survey of all practicing veterinarians. An equine showing signs of fever, deranged consciousness, depression, motor irritation, and paralysis in the absence of any other disease or condition was recorded as a case of encephalomyelitis. This survey is conducted annually by the Veterinary Public Health Section of the Iowa State Department of Health.

RESULTS AND DISCUSSION

A total of 1,037 bird bloods representing 41 species was collected from June through September and tested for SLE and WEE antibodies. Birds were trapped in three Iowa cities (Ames, Davenport, and Des Moines) as part of a surveillance program for arbovirus activity. The number of birds tested for the presence of HI antibodies against WEE virus is listed in Table 1 by location and month collected. As expected, the numbers of birds with antibody titers of 1:20 or greater increased throughout the arbovirus season. The greatest amplification of WEE virus occurred in late summer. In Des Moines, a major population area, WEE antibodies in wild birds rose from 0 in June to nearly 10%. Of the total number tested, 23 (19 juvenile, 4 adults) demonstrated WEE antibody (Table 1).

Table 2 lists by location and month of collection the results of HI testing of birds for SLE antibody. Only 4 adult house sparrows were positive for SLE antibody. These sparrows had been trapped in Des Moines, an area thought to be endemic for SLE virus. None of the birds trapped in Ames, or Davenport had SLE antibody at a serum dilution of 1:20. These results are compatible with virus isolation data because SLE virus was not recovered from any of the mosquitoes tested.

Fig. 1 shows the geographic distribution of encephalomyelitis in horses during 1977. There were 229 cases of equine encephalomyelitis, with 57 deaths reported. These cases occurred in 62 of Iowa's 99 counties and were distributed throughout the state. The greatest number of cases occurred in Lee County

Table 1. Birds with hemagglutination-inhibition antibodies (1:20 or greater) to western equine encephalomyelitis virus by location and month of collection, Iowa, 1977.

Date	Number (Percent) with HI Titer ≥20/No. Tested			
	Ames	Davenport	Des Moines	Totals
June	0/78	0/6	0/120	0/204
July	3[a]/190 (1.6)	1[Δ]/69 (1.4)	—	4/259 (1.5)
August	7[c]/159 (4.4)	0/67	1[a]/114 (0.9)	8/340 (2.4)
September	0/6	2[b]/91 (2.2)	5[a]/52 (9.6)	7/149 (4.7)
October	—	1[d]/49 (2.0)	3[a](36 (8.3)	4/85 (4.7)

[Δ] Adult rock dove.
[a] Juvenile house sparrows.
[b] One adult and one juvenile house sparrow.
[c] Six juvenile house sparrows and one adult brown thrasher.
[d] Adult house sparrow.

where 14 cases and 3 deaths were recorded. Polk County, an area where nearly 10% of the birds surveyed were positive for WEE, had 8 reported cases of equine encephalomyelitis, with 3 deaths. Warren County, bordering Polk County to the south, had 13 cases of equine encephalomyelitis and 3 deaths. Table 3 lists the number of cases of encephalomyelitis in horses from 1973 to 1977. Although the total number of cases was greatest in 1975, the percentage of deaths recorded that year (21.7) was less than the average (24.0) for the 5-year period.

greatest number of laboratory-confirmed cases of mosquito-borne encephalitides occurred in 1975. That year, the 36 cases included: 12 cases of LAC, 19 SLE and 5 WEE. In 1976, there were 5 cases of LAC and 3 cases of SLE. Three cases of LAC and 1 case of WEE represent the total number of cases of encephalitis of arbovirus etiology in 1977. As expected, the 3 cases of LAC in 1977 were children. The ages of these children were 2½, 12, and 16 years. The single case of WEE, confirmed by serologic examination, occurred in a 58-year-old

Table 2. Birds with hemagglutination-inhibition (HI) antibodies (1:20 or greater) to St. Louis encephalitis virus by location and month of collection, Iowa, 1977.

Date	Number (Percent) with Titers ≤20/No. Tested			
	Ames	Davenport	Des Moines	Totals
June	0/78	0/6	2[a]/120 (1.7)	2/204 (1.0)
July	0/190	0/69	—	0/259
August	0/159	0/67	1[a]/114 (0.9)	1/340 (0.3)
September	0/6	0/91	0/52	0/149
October	—	0/49	1[a]/36 (2.8)	1/85 (1.2)

[a] adult house sparrows.

There were 4 human cases of mosquito-borne encephalitis in Iowa in 1977 on the basis of serologic results obtained when paired sera were tested for HI, CF, and neutralizing antibodies. Table 4 lists the laboratory-confirmed cases of LAC, SLE, and WEE for the 12-year period from 1965 to 1977. The male farmer and horse owner living in Howard County in northeastern Iowa.

During the summer of 1977, 26,939 mosquitoes were collected in CO_2-baited CDC light traps, identified by species, and divided into 634 pools for arbovirus studies. Table 5 lists the major species found and the 73 arboviruses isolated from these

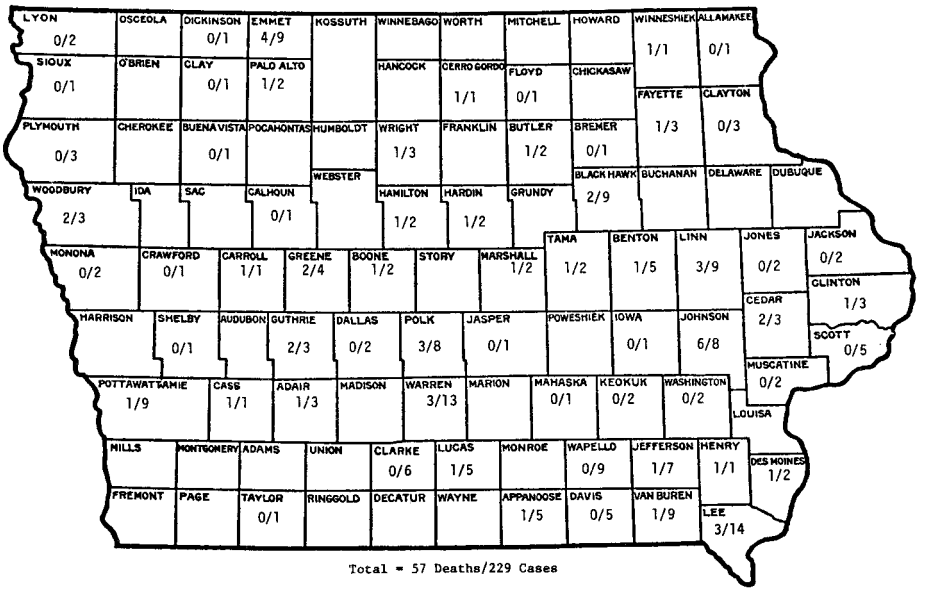

Fig. 1. Geographic distribution of encephalomyelitis in horses during 1977.

species. Cache Valley (CV), 21 Flanders (FL), 3 Turlock (Tur), 37 trivittatus (TVT), and 11 WEE viruses were identified. The single CV isolate was from a pool of 37 *Anopheles* species collected in mid-August in Davenport, Iowa. This virus has been previously isolated in our laboratory from *Aedes nigromaculis, An. punctipennis, Culiseta inornata,* and *Coquillettidia perturbans* mosquitoes (Wong et al. 1973, 1978).

Western equine encephalomyelitis virus was isolated from 2 pools of *Ae. trivittatus,* 4 pools of *Cx. pipiens* complex mosquitoes, and 5 pools of *Cx. tarsalis* mosquitoes. WEE virus usually is isolated from mid-July through August, but one isolate was made from a pool of *Cx. pipiens* complex mosquitoes trapped on the 17th of June 1977. This early isolate of WEE

Table 3. Clinically diagnosed cases of encephalomyelitis in horses in Iowa 1973–1977.

Year	Cases	Deaths	Percent (Death/Cases)
1973	95	27	28.4
1974	154	40	26.0
1975	434	94	21.7
1976	41	8	19.5
1977	229	57	24.8

Table 4. Mosquito-borne encephalitis in Iowa 1965–1977.

Year	La Crosse	Western Equine	St. Louis	Total
1965	6	—	—	6
1966	—	—	—	—
1967	11	—	—	11
1968	4	—	—	4
1969	7	—	—	7
1970	14	1	—	15
1971	3	—	2	5
1972	1	—	—	1
1973	5	—	—	5
1974	2	—	—	2
1975	12	5	19	36
1976	5	—	3	8
1977	3	1	—	4
Total	73	7	24	104

Table 5. Arboviruses isolated from mosquitoes in Iowa, 1977.

Species	Number Collected (Pools)	Arbovirus Isolates
Ae. triseriatus	242(19)	
Ae. trivittatus	8,707(179)	Trivittatus (36) WEE (2)
Ae. vexans	2,820(62)	
An. punctipennis	1,019(39)	
Cx. pipiens complex*	10,390(221)	Flanders (13) WEE (4) Turlock (2)
Cx. tarsalis	3,441(84)	Flanders (8) WEE (5) Turlock
Other species	320(30)	Cache Valley Trivittatus
Total	26,939(634)	73

* *Cx. pipiens* complex—*Cx. pipiens pipiens, Cx. pipiens quinquefasciatus, Cx. restuans* and *Cx. salinarius.*

virus is predictable, however, because WEE cases, although sporadic throughout the state, can occur in late May and early June. This is interesting inasmuch as the data indicated that none of the 204 birds tested in June had WEE antibody (Table 1).

TVT continues to be the most frequently isolated virus in mosquitoes in Iowa. Unlike the other mosquito-borne viruses, which are isolated only during a portion of the "mosquito season," TVT was isolated every month in which mosquitoes were collected. From the 4th of June through the 23rd of September, TVT virus was recovered from 36 different pools of *Ae. trivittatus* and one pool of *Aedes* species of mosquitoes. This virus probably overwinters in the eggs of *Ae. trivittatus* (Andrews et al. 1977; Christensen et al. 1978) The role of TVT virus as an etiologic agent of disease has yet to be elucidated.

Mosquito populations were studied in 8 communities located throughout Iowa as an index for control measures. Table 6 lists the 21 species collected in standard New Jersey light traps. A total of 151,530 mosquitoes were collected in 1977 versus 39,946 mosquitoes collected in 1976. *Ae. vexans* was the predominant species, constituting 47.6% of the total collected (Fig. 2). There was nearly a 3-fold increase in

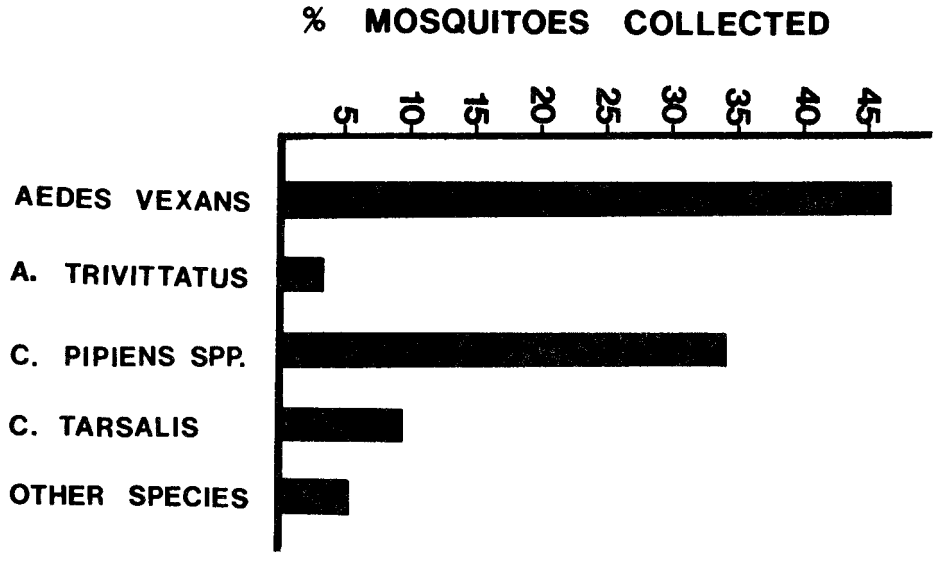

Table 6. Mosquito species collected in Iowa communities in 1977.

Species	Ames	Des Moines	Waterloo	Iowa City	Council Bluffs	Sioux City	Daven-port	Dubuque	Total	% Total
Aedes species	194	33	3	80	224	66	9	47	656	.43
Ae. campestris	—	—	—	11	—	—	—	—	11	.01
Ae. canadensis	—	—	—	3	—	—	—	—	3	
Ae. dorsalis	1	1	—	2	11	8	—	—	23	.02
Ae. nigromaculis	67	46	11	31	379	436	57	3	1,030	.68
Ae. spenceri	—	—	—	—	3	—	—	—	3	
Ae. triseriatus	10	4	1	19	1	—	2	11	37	.03
Ae. trivittatus	2,078	67	33	133	606	23	68	1,655	4,663	3.08
Ae. vexans	20,371	9,882	11,966	11,492	5,063	678	9,292	3,494	72,238	49.68
Ae. sollicitans	1	1	—	1	23	3	9	—	38	.03
Anopheles punctipennis	2,084	746	154	374	66	38	511	120	4,093	2.70
An. quadrimaculatus	1	5	1	2	3	1	5	2	20	.01
An. walkeri	—	—	—	—	—	—	—	1	1	34.24
Culex pipiens complex	31,171	3,452	1,855	4,066	1,878	911	4,760	1,847	51,880	9.27
Cx. tarsalis	13	1,833	563	458	1,405	886	944	745	14,047	.65
Culiseta inornata	433	154	85	76	61	27	125	25	986	
Coquillettidia perturbans	1	—	—	—	1	—	—	—	2	
Orthopodomyia signifera	—	—	—	1	—	—	—	1	2	
Psorophora ciliata	2	—	—	2	3	—	2	—	9	
Ps. horrida	5	2	—	—	—	—	—	—	7	
Ps. signipennis	1	—	—	—	21	1	2	—	25	.02
Uranotaenia sapphirina	39	16	14	42	—	—	34	2	147	.10
Unidentifiable specimens	513	72	81	476	288	48	68	63	1,609	1.06
Total	66,185	16,314	14,767	17,209	10,036	3,126	15,888	8,005	151,530	100.00
% Total	43.68	10.77	9.75	11.36	6.62	2.06	10.49	5.28	100	

the numbers of *Cx. pipiens* complex mosquitoes collected in 1977 (51,880) compared with 1976 (17,697). The increase in the numbers of these mosquitoes is cause for concern because these numbers may predict increases in mosquito populations for the coming season.

CONCLUDING REMARKS

Iowa recorded 4 human cases of mosquito-borne encephalitis in 1977. LAC clearly is the most significant arbovirus in Iowa, with cases occurring annually since 1967. The presence of WEE in mosquitoes, amplification of this virus in birds, and the number of cases of equine encephalomyelitis that occurred in 1977, along with the paucity of human cases, is not fully understood. The absence of SLE activity in mosquitoes, birds, and man is surprising in light of the unusually large populations of *Culex* mosquitoes present in 1977.

References Cited

Andrews, W. J., Rowley, W. A., Wong, Y. W., Dorsey, D. C., and Hausler, W. J., Jr. 1977. Isolation of trivittatus virus from larvae and adults reared from field-collected larvae of *Aedes trivittatus* (Diptera: Culicidae). J. Med. Entomol. 13:699–701.

Christensen, B. M., Rowley, W. A., Wong, Y. W., Dorsey, D. C. and Hausler, W. J., Jr., 1978. Laboratory studies of transovarial transmission of trivittatus virus by *Aedes trivittatus*, Am. J. Trop. Med. Hyg. 27(1): 184–86.

Holden, P., Muth, D. and Shriner, R. B. 1966. Arbovirus hemagglutination-inhibitor in avian sera: Inactivation with protamine sulfate. Am. J. Epidemio. 84:67–73.

Lennette, E. H. and Schmidt. N. J. 1969. Diagnostic procedures for viral and rickettsial infections, 4th Ed., pp. 124–133. Am. Public Health Assoc., New York, N.Y.

Rowley, W. A., Wong, Y. W., Dorsey, D. C. and Hausler W. J., Jr., 1973. Field studies on mosquito-arbovirus relationships in Iowa, 1971. J. Med. Entomol. 10:613–617.

Sudia, W. D. and Chamberlain, R. W., 1967. Collection and processing of medically important arthropods for arbovirus isolation. Center for Disease Control, Public Health Serv., USDHEW, Atlatnta, Georgia, 65 pp.

U.S. Public Health Service. 1965. Standardized diagnostic complement fixation method and adaptation to micro test. Public Health Monograph No. 74. U.S. Public Health Serv. Publication 1228, U.S. Gov. Printing Off. Washington, D.C., 34 pp.

Wong, Y. W., Dorsey, D. C., Humphreys, M. J. and Hausler, W. J., Jr., 1970. Arbovirus encephalitis surveillance in Iowa. Health Lab. Sci. 7:117–123.

Wong, Y. W., Rowe, J. A., Dorsey, D. C., Humphreys, M. J. and Hausler, W. J., Jr. 1971. Arboviruses isolated from mosquitoes collected in southeastern Iowa in 1966. Am. J. Trop. Med Hyg. 20:726–729.

Wong, Y. W., Rowley, W. A., Dorsey, D. C. and Hausler, W. J., Jr., 1978. Surveillance of arbovirus activity in Iowa during 1972–1975. Mosquito News 38:245–251.

Wong, Y. W., Rowley, W. A., Rowe, J. A., Dorsey, D. C., Humprhreys, M. J. and Hausler, W. J., Jr., 1973. California encephalitis studies in Iowa during 1969, 1970, and 1971. Health Lab. Sci. 10:88–95.

ERRATUM

In the Table of Contents for the September number of *Mosquito News* the name of the senior author of the article, "Jamestown Canyon Virus in Connecticut," is incorrectly listed as H. F. Sprague. This should read H. F. Sprance.