

ARBOVIRUS SURVEILLANCE IN CONNECTICUT. II. CALIFORNIA SEROGROUP¹

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ABSTRACT. Fifteen of 19 isolations of Jamestown Canyon virus from Connecticut mosquitoes tested since 1969 were recovered from *Aedes abserratus*; the minimum field infection rate (MFIR) for the 10-year period was 1:438. The other 4 strains were from *Ae. cantator* (2 isolates) (MFIR 1:4024), *Ae. vexans*

(MFIR 1:25353), and *Coquillettidia perturbans* (MFIR 1:26666).

Keystone virus is reported for the first time in New England. One strain was recovered from *Ae. aurifer* and another from *Chrysops obsoletus*. The latter isolation is the first published record of this virus from a tabanid.

INTRODUCTION

The first isolate of a California group virus in Connecticut was from a pool of 20 adult female *Aedes abserratus* from light trap collections in the town of Simsbury between 27 June and 7 July 1966 (Whitman et al. 1968). This strain was subsequently identified as Jamestown Canyon virus and 4 additional isolates were reported—1 from *Ae. cantator* (1973), 1 from *Ae. vexans* (1975), and 2 from *Ae. abserratus* (1975) (Sprance et al. 1978).

The present paper reports an additional 15 isolates of Jamestown Canyon virus, 2 isolates of Keystone virus from mosquitoes and deer flies, and summarizes the results of 10 years (1969–1978) of arbovirus surveillance for California encephalitis group viruses in Connecticut.

MATERIALS AND METHODS

Most of the procedures used in these studies were described in detail in part I (Main et al. 1979) except for the serologic

techniques used to type the California encephalitis group isolates. Complement-fixation (Sprance and Shope 1977) and plaque-reduction neutralization tests in Vero cells (Buckley 1975) with single inoculation hamster sera were employed for serotyping.

RESULTS

Nineteen isolates of Jamestown Canyon virus were recovered from mosquitoes in Connecticut since 1969 (Table 1). Fifteen of the strains were from *Ae. abserratus*; the minimum field infection rate (MFIR) in this species for the 10-year period was 1 positive for every 438 tested. Virus was isolated from *Ae. abserratus* every year from 1975 through 1978; in the 6 years prior to that date, fewer than 300 *Ae. abserratus* were tested (Table 2). Infection rates varied from 1:106 in 1975 to 1:744 in 1976. Four strains of this virus were isolated from other mosquito species: 2 from *Ae. cantator* (MFIR 1:4024) and 1 each from *Ae. vexans* (MFIR 1:25353) and *Coquillettidia perturbans* (MFIR 1:26666) (Table 1).

The earliest Jamestown Canyon virus isolation was from mosquitoes collected in May; most isolations were from June and a few from July. Virus was recovered

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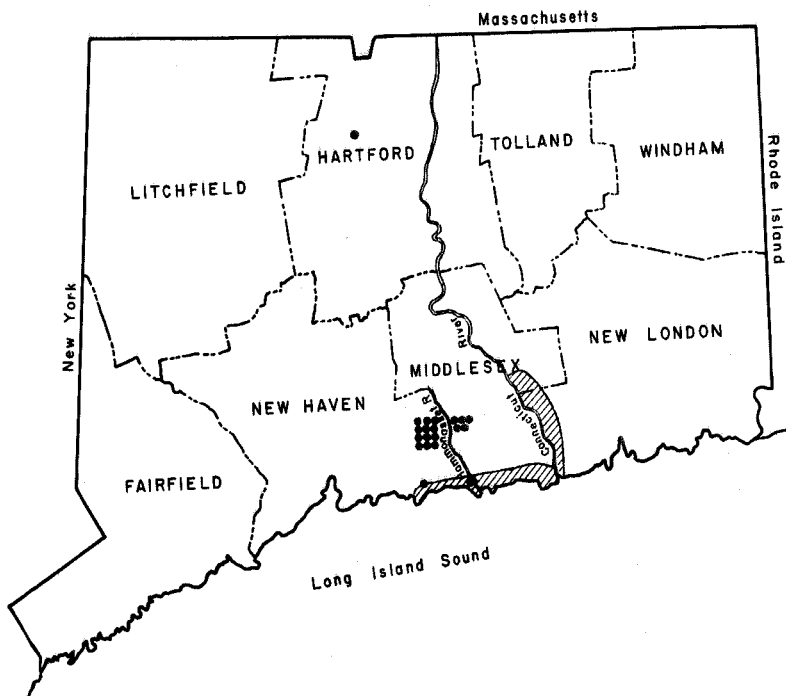


Figure 1. The locations of California group arbovirus isolations from mosquitoes in Connecticut, 1969 through 1978.

Legend • Jamestown Canyon virus.

//// Keystone virus.

from mosquitoes in areas of southcentral Connecticut where the biggest effort was made to collect *Ae. abserratus* (Fig. 1).

Keystone virus was isolated on two occasions (Table 1). In 1975, this virus was recovered from a pool of 63 adult female *Ae. aurifer* collected in four coastal towns in southern Connecticut. In 1976, the virus was again isolated, this time from a pool of 59 adult female *Chrysops obsoletus* from three towns along the Connecticut River (Fig. 1). Very few specimens of either species were collected or tested (Table 2); therefore, conclusions about the infection rates, seasonal prevalence, distribution, or significance of these isolates cannot be made. Both isolates were identified by complement-fixation and plaque-reduction neutralization tests (Table 3).

California group viruses were not recovered from 52,560 adult female mosquitoes other than *Aedes* or *Coquillettidia* species, from 13,767 adult male mosquitoes including 5810 *Aedes* spp. or from 9791 mosquitoes reared from field collected larvae including 9742 *Aedes* spp. (Table 4). Except for the single Keystone isolate from *Ch. obsoletus*, virus was not detected in 4545 Tabanidae (*Chrysops* spp., *Hybomitra* spp., *Tabanus* spp.), 1999 Simuliidae (*Cnephia* spp., *Prosimulium* spp., *Simulium* spp.), 39,673 Ceratopogonidae (*Culicoides* spp.), or 665 Rhagionidae (*Symphoromyia* spp.).

DISCUSSION

Eighty percent of the Jamestown Canyon virus isolations in Connecticut were

from *Ae. abserratus*. Infection rates in this species were 10- to 60-fold greater than in the other 3 species from which this virus was recovered, suggesting that *Ae. abserratus* could be an important vector of Jamestown Canyon virus at least in Connecticut. However, confirmation must await the demonstration of transmission either in the laboratory or in the field.

Ae. abserratus is a very abundant univoltine species throughout New England. Larvae begin hatching in late March or early April, depending upon meteorological conditions in southern New England; adults may appear as early as late April and remain through late July, although the population peak is reached in early June (Wallis 1960, Main et al. 1968). This species is an important pest of man and domestic animals.

Mammals, particularly larger species including horses, cattle, and deer, are preferred hosts (Magnarelli 1977).

The distribution of *Ae. abserratus*—northeastern United States and southeastern Canada (Knight and Stone 1977)—does not coincide with that of Jamestown Canyon virus (including Jerry Slough and South River varieties) (Sudia et al. 1971, Parkin et al. 1972, Berge 1975) indicating that other vectors are also involved. The largest number of isolates reported in the literature (Sudia et al. 1971, Parkin et al. 1972, Wills et al. 1974, LeDuc et al. 1975, Berry et al. 1977) were from *Ae. communis* group mosquitoes. This group includes *Ae. abserratus* in Wisconsin, where most of the *Ae. communis* group isolates were reported (DeFoliart et al. 1969), but *Ae. abserratus* is not

Table 1. California group virus isolations from Diptera collected in Connecticut, 1969-1978.

Strain	Species	Pool Size*	Location	Date	Serotype
73660	<i>Aedes cantator</i>	50	Guilford	VII.73	JC
Ar-74-75	<i>Aedes abserratus</i>	41	Madison	17-20.VI.75	JC
Ar-78-75	<i>Aedes vexans</i>	25	Madison	16-20.VI.75	JC
Ar-83-75	<i>Aedes abserratus</i>	25	Killingworth	19-29.VI.75	JC
Ar-623-75	<i>Aedes aurifer</i>	63	Westbrook, Guilford, Clinton & Madison	16.VI-13.VIII.75	KEY
Ar-64-76	<i>Aedes abserratus</i>	33	Killingworth	2-3.VI.76	JC
Ar-505-76	<i>Coquillettidia perturbans</i>	100	Killingworth	1-2.VII.76	JC
Ar-1274-76	<i>Chrysops obsoletus</i>	59	Lyme, Old Lyme, & East Haddam	1-28.VII.76	KEY
Ar-47-77	<i>Aedes abserratus</i>	50	Madison	2-3.VI.77	JC
Ar-99-77	<i>Aedes abserratus</i>	50	Killingworth	24-25.V.77	JC
Ar-108-77	<i>Aedes abserratus</i>	48	Killingworth	2-3.VI.77	JC
Ar-111-77	<i>Aedes abserratus</i>	43	Madison	13-16.VI.77	JC
Ar-112-77	<i>Aedes abserratus</i>	50	Madison	15.VI-1.VII.77	JC
Ar-114-77	<i>Aedes abserratus</i>	46	Madison	13-16.VI.77	JC
Ar-392-77	<i>Aedes cantator</i>	46	Madison	16.VI-7.IX.77	JC
Ar-208-78	<i>Aedes abserratus</i>	25	Madison	8-9.VI.78	JC
Ar-213-78	<i>Aedes abserratus</i>	32	Madison	14-15.VI.78	JC
Ar-315-78	<i>Aedes abserratus</i>	25	Madison	5-6.VI/78	JC
Ar-317-78	<i>Aedes abserratus</i>	25	Madison	5-6.VI.78	JC
Ar-321-78	<i>Aedes abserratus</i>	25	Madison	5-6.VI.78	JC
Ar-326-78	<i>Aedes abserratus</i>	25	Madison	5-6.VI.78	JC

* All adult females.

JC=Jamestown Canyon Virus.

KEY=Keystone Virus.

Table 2. Adult female Diptera from Connecticut tested for arbovirus infections, 1969-1978.

Species	1969	1970	1971	1972	1973	1974	1975	1976	1977	1978	Totals
<i>Aedes abstrerratus</i>	0	91	153	2	1	27	212	744	2185	3149	6564
<i>Aedes aurifer</i>	0	7	9	0	0	393	149	51	6	0	615
<i>Aedes canadensis</i>	809	841	351	153	43	760	2775	12681	3975	4	22392
<i>Aedes cantator</i>	3	0	0	38	1606	456	1253	4046	645	0	8047
<i>Aedes cinereus</i>	441	772	390	108	16	70	94	2444	1007	2	5344
<i>Aedes communis</i>	0	1	14	0	0	0	0	0	0	0	15
<i>Aedes intrudens</i>	0	4	5	0	0	0	0	0	0	0	9
<i>Aedes sollicitans</i>	0	0	0	1	188	141	617	483	45	0	1475
<i>Aedes sticticus</i>	0	0	0	0	0	0	2	6	0	0	8
<i>Aedes stimulans</i> group*	1	855	219	0	28	66	275	780	796	3	3023
<i>Aedes provocans</i>	0	0	1	0	0	0	0	0	0	0	1
<i>Aedes triseriatus</i>	290	137	34	19	91	432	665	796	202	2211	4877
<i>Aedes trivittatus</i>	50	0	0	0	0	0	216	300	62	0	628
<i>Aedes vexans</i>	3900	784	2313	321	3824	1008	5381	7515	307	0	25353
<i>Aedes</i> species unknown	0	57	3	9	0	0	0	0	0	0	69
<i>Coquillettidia perturbans</i>	1	214	6	12	1165	1587	2519	19197	1963	2	26666
<i>Culex</i> species	5812	1655	1483	1486	1348	2158	2455	4862	591	2852	24702
<i>Culiseta</i> species	3228	2257	1981	3063	955	405	1826	3145	993	3793	21646
<i>Psorophora</i> species	130	0	0	0	3	0	35	75	0	0	243
<i>Uranotaenia sapphirina</i>	331	125	154	59	36	218	313	546	122	0	1904
<i>Orthopodomyia signifera</i>	0	0	0	0	2	0	1	1	0	0	4
<i>Anopheles</i> species	743	703	94	808	48	120	260	635	649	1	4061
TOTAL CULICIDAE	15739	8503	7210	6079	9354	7841	19048	58307	13548	12017	157646
<i>Chrysops obsoletus</i>	0	0	0	0	0	0	0	88	0	0	88
Other <i>Chrysops</i> species	0	0	0	0	0	0	2590	1551	25	1	4167
<i>Hybomitra</i> species	0	0	0	0	0	0	15	152	11	1	179
<i>Tabanus</i> species	0	0	0	0	0	0	47	38	0	0	85
TOTAL TABANIDAE	0	0	0	0	0	0	2652	1829	36	2	4519
<i>Cnephia mutata</i>	0	0	0	0	0	0	0	20	1	0	21
<i>Prosimulium</i> species	0	0	0	0	0	0	0	1103	21	0	1124
<i>Simulium</i> species	0	0	0	0	0	0	339	488	27	0	854
TOTAL SIMULIIDAE	0	0	0	0	0	0	339	1611	49	0	1999
<i>Culicoides</i> species	0	0	0	0	0	0	0	39477	196	0	39673
TOTAL GERATOPOGONIDAE	0	0	0	0	0	0	0	39477	196	0	39673

Table 2. Continued.

Species	1969	1970	1971	1972	1973	1974	1975	1976	1977	1978	Totals
<i>Symphoromyia</i> species	0	0	0	2	0	0	0	663	0	0	665
TOTAL RHAGIONIDAE	0	0	0	2	0	0	0	633	0	0	665
TOTAL DIPTERA	15739	8503	7210	6081	9354	7841	22039	101887	13829	12019	204502

* Includes *Aedes excrucians*, *Aedes fitchii*, and *Aedes stimulans*.

known to occur in Alberta, where this virus was also recorded from the *communis* complex (Iversen et al. 1969).

Jamestown Canyon virus has been reported from at least 9 species of *Aedes* in addition to the *Ae. communis* group, 2 species each of *Psorophora*, *Culex*, *Culiseta*, and *Hybomitra* and 1 species each of *Anopheles*, *Coquillettidia*, and *Chrysops* (Sudia et al. 1971, Parkin et al. 1972, Wills et al. 1974, LeDuc et al. 1975, Berry et al. 1977). However, there are relatively few isolations from any of these species and the significance of these isolations is unknown. Of particular interest is the isolate from *Ae. triseriatus* reared from eggs collected in Ohio (Berry et al. 1977). In our studies, virus was not detected in 8317 *Ae. triseriatus* reared from larvae or 5081 collected as adults in the same locations as infected *Ae. aberratus* adults.

Information about the vertebrate host is not available from our studies in Connecticut. Deer are considered to be an important host for Jamestown Canyon virus in California (Emmons 1968), Texas (Issel et al. 1973), and Wisconsin (Issel et al. 1972; Issel 1973, 1974). Thompson (in Sudia et al. 1971) reported evidence suggesting that this virus was the etiologic agent of a mild febrile illness in forest workers in Wisconsin. Neutralizing antibody was detected in 71% of 325 rural residents from Alaska (Feltz et al. in Henderson and Coleman 1971).

Keystone virus is reported here for the 1st time in New England. More than 80% of the 460 mosquito isolations of this virus summarized by Sudia and his colleagues (1971) were from the *Ae. atlanticus/tormentor* complex, 11% from *Ae. infirmatus*, and 5% from *Ae. taeniorhynchus*. Only the latter of these species occurs in New England, and it was encountered only occasionally in our studies; specimens were not tested for virus infections. Keystone virus was reported from *Anopheles punctipennis* in Pennsylvania (Wills et al. 1974) and from *Ae. canadensis* as well as *Ae. atlanticus* on the Del Mar Va Peninsula (LeDuc et al. 1975). This virus has been associated with cotton rats (Sig-

Table 3. Serological identification of two strains of Keystone virus from Connecticut.

Virus (strain)	Antigen				Ascitic Fluid*	
	Ar-623-75		Ar-1274-76		Ar-623-75	Ar-1274-76
	CF	NT	CF	NT	CF	NT
Keystone (B64-5587)	64/64***	64/≥64	64/64	≥64/≥64	256/128	512/256
California encephalitis (BFS 283)	<8/64	<8/≥64	<8/64	<8/≥64	64/128	64/256
San Angelo (USA 20230)	<8/≥256	128/≥256	<8/≥256	128/≥256	64/128	128/256
LaCrosse (original)	16/128	<8/≥128	16/128	<8/≥128	64/128	64/256
Snowshoe Hare (original)	<8/64	<8/≥64	<8/64	<8/≥64	16/128	8/256
Jamestown Canyon (Ar-83-75)	<8/64	8/≥64	<8/64	<8/≥64	32/128	128/256
South River (NJO-94-5)	<8/32	<8/≥64	8/32	<8/≥64	64/128	64/256
Trivittatus (USA 993)	<8/≥256	<8/≥	<8/≥256	<8/≥64	32/128	16/256
Melao (Tr 9375)	<8/16	8/≥64	<8/16	8/≥64	64/128	128/256
Serro do Navio (BeAr 103645)	<8/8	<8/≥64	16/8	<8/≥64	128/128	128/256
Tahyna (Cz 92)	16/128	16/≥128	8/128	≥32/≥128	64/128	128/256
Inkoo (KN 3641)	16/64	8/≥64	16/64	8/≥64		

* Ar-623-75 and Ar-1274-76 hyperimmune mouse ascitic fluids.

** Prototype strains—single inoculation hamster sera.

*** Heterologous serum titer reciprocal or log neutralization index/homologous serum titer reciprocal or log neutralization index.

CF=Complement-fixation test.

NT=Plaque-reduction neutralization test in Vero cells.

modon hispidus) in the southern states (Sudia et al. 1971, Taylor et al. 1971), but this species does not occur in New England.

The isolate from *Ch. obsoletus* is the 1st report of Keystone virus from a species of Tabanidae, although both Jamestown Canyon (DeFoliart et al. 1969) and LaCrosse (Wright et al. 1970) viruses have been recovered from *Chrysops* and/or *Hybomitra* species in Wisconsin. The significance of these findings is unknown at present.

Although Keystone virus has not been associated with human disease, antibody was detected in 16 to 21% of sera from residents of Louisiana (Henderson and Coleman 1971) and Florida (Wellings in Parkin et al. 1972).

Other California encephalitis group viruses were not encountered in the present survey, although several are

known to occur in neighboring states. LaCrosse, trivittatus, and snowshoe hare serotypes may eventually be detected in Connecticut (Sudia et al. 1971).

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Table 4. Immature and adult male Diptera from Connecticut tested for arboviruses,* 1974-1978.

Species	Immatures**						Males			
	1974	1975	1976	1977	1978	Total	1976	1977	1978	Total
<i>Aedes abserratus</i>	1	13	3	0	176	193	7	23	37	67
<i>Aedes awrifer</i>	0	0	0	0	0	0	2	0	0	2
<i>Aedes canadensis</i>	25	60	822	0	0	907	1286	48	0	1334
<i>Aedes cantator</i>	0	0	0	0	0	0	109	5	0	114
<i>Aedes cinereus</i>	50	56	21	0	0	127	1983	50	0	2033
<i>Aedes communis</i>	8	0	3	0	0	11	0	0	0	0
<i>Aedes excrucians</i>	46	0	16	0	0	62	0	0	0	0
<i>Aedes fitchii</i>	2	0	0	0	0	2	0	0	0	0
<i>Aedes stimulans</i>	1	0	110	0	0	111	0	0	0	0
<i>Aedes stimulans</i> group***	0	0	0	0	0	0	72	30	0	102
<i>Aedes sollicitans</i>	0	0	0	0	0	0	5	0	0	5
<i>Aedes provocans</i>	12	0	0	0	0	12	0	0	0	0
<i>Aedes triseriatus</i>	50	8073	188	6	0	8317	194	10	0	204
<i>Aedes trivittatus</i>	0	0	0	0	0	0	7	0	0	7
<i>Aedes vexans</i>	0	0	0	0	0	0	1794	3	0	1797
<i>Aedes</i> species unknown	0	0	0	0	0	0	0	145	0	145
<i>Coquillettidia</i> <i>perturbans</i>	0	0	0	0	0	0	93	48	2	143
<i>Culex</i> species	0	0	36	0	0	36	4284	158	3	4445
<i>Culiseta</i> species	2	0	11	0	0	13	2496	379	37	2912
<i>Psorophora ferox</i>	0	0	0	0	0	0	3	0	0	3
<i>Uranotaenia sapphirina</i>	0	0	0	0	0	0	310	77	0	387
<i>Anopheles</i> species	0	0	0	0	0	0	38	29	0	67
<i>Chrysops geminatus</i>	0	0	0	0	0	0	0	3	0	3
<i>Tabanus pumilus</i>	0	0	0	0	0	0	23	0	0	23
<i>Culicoides</i> species	0	152	0	0	0	152	0	0	0	0
Total Diptera	197	8354	1210	6	176	9943	12706	1008	79	13793

* All negative.

** Reared in the laboratory and tested as adults.

*** Includes *Aedes excrucians*, *Aedes fitchii*, and *Aedes stimulans*.

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