THE RECURRENCE OF ST. LOUIS ENCEPHALITIS VIRUS IN *CULEX NIGRIPALPUS* MOSQUITOES IN JAMAICA, 1963

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St. Louis encephalitis virus (SLE) was first isolated in Jamaica from *Culex nigripalpus* Theobald collected at the Caymanas Sugar Plantation near Kingston in June of 1962 (Belle *et al.* 1964). Later that same year, 40 additional SLE virus isolations were made from the same species during an epidemic in the Tampa Bay area of Florida (Chamberlain, *et al.* 1964; Dow, *et al.* 1964). Experimental infection and transmission studies with Florida strains of *C. nigripalpus* and SLE virus showed the species to be exceptionally susceptible, with an infection rate of 100 percent and a transmission rate of nearly 100 percent (Sudia and Chamberlain, 1964). On the basis of the above studies, *C. nigripalpus* was firmly established as a vector of SLE in these areas.

In the summer of 1963, the University of West Indies and the Communicable Disease Center (CDC), U. S. Public Health Service, collaborated in the investigation of a dengue epidemic in Kingston, Jamaica. During entomological studies of this epidemic, CDC battery-powered light traps (Sudia and Chamberlain, 1962) were operated briefly at the University of W. I. study areas which had yielded the SLE isolation the year before. The collections were made primarily to determine the efficiency of the CDC light trap under Jamaican conditions.

MATERIALS AND METHODS. Eight CDC battery-operated light traps were run for two nights on the Caymanas Sugar Plantation on August 30 and 31, 1963. Four traps each were also used for a single night at the Dalvey and Holland Bay study sites. In addition, a few mosquitoes were collected from donkey-baited traps located nearby. The specimens were shipped on dry ice by air to the Arbovirus Vector Laboratory, CDC, Atlanta, Georgia, for virus isolation studies. Procedures for mosquito trapping, handling, and virus testing were the same as those already reported (Chamberlain *et al.* 1964).

Preliminary identification of isolates was by means of the complement fixation test (Hammon and Work, 1964). Crude borate saline antigen was prepared for each isolate and tested with hyperimmune mouse sera against the following viruses: Eastern, Western, St. Louis, and California encephalitis, Ilheus, Tensaw, yellow fever, and dengue strains, I, II, III and IV.

Cross neutralization tests in weanling mice were performed to confirm the tentative identifications. Heterologous and homologous cross-neutralization tests were conducted employing hyperimmune ascitic fluids and hyperimmune mouse or rabbit sera against the isolates and SLE-TBH-28, SLE-Parton, Ilheus and Dengue II viruses. Ten-fold dilutions of infected suckling mouse brain suspensions were mixed with equal parts of test serum and incubated for two hours in a 37°C water bath; then 0.03 ml aliquots were inoculated intracerebrally into each of six weanling mice per dilution. Inoculated mice were observed for two weeks and neutralization indices calculated according to the methods of Reed and Meunch (1938).

RESULTS AND DISCUSSION. A total of 1145 mosquitoes, representing 13 species, were collected in 24 light trap nights of operation (table 1) at the three locations. *C. nigripalpus* was by far the predominant mosquito attracted to the light traps, accounting for 925 of the 947 mosquitoes captured at the Caymanas site. In the donkey-baited trap, used the first night of collection at Caymanas, only ten mosquitoes were captured.

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The light-trapped mosquitoes were tested in suckling mice in 41 pools; two of the pools of *C. nigripalpus* from the Caymanas Estates yielded viral agents.

Crude borate saline suspensions of mouse brains infected with these two isolates (J3-16h and J3-16n) reacted only with known SLE anti-sera (table 2), indicating that the isolates were closely related to SLE virus. Subsequent cross-neutralization tests demonstrated the identity of the isolates as SLE virus (table 3). Hyperimmune ascitic fluid and sera prepared against the two Jamaican isolates significantly neutralized both the Jamaican isolates and known strains of SLE virus,

TABLE 1.—Live mosquito catches made in Jamaica August 30–September 1, 1963, with CDC battery-operated light traps.

Mosquito species	Caymanas Estates 8/30–31/63	Holland Bay 9/1/63	Dalvey 9/1/63	Totals
Aedes scapularis	2* / (1)**	..	..	2 / (1)
taeniorhynchus	..	27 / (1)	32 / (1)	59 / (2)
tortilis	8 / (2)	..	..	8 / (2)
Anopheles albimanus	..	1 / (1)	..	1 / (1)
grabhamii	7 / (2)	1 / (1)	10 / (1)	18 / (4)
Culex corniger	2 / (2)	..	..	2 / (2)
(Melanoconion) sp.	..	2 / (1)	22 / (2)	24 / (3)
nigripalpus	925*** / (18)	7 / (1)	6 / (1)	938 / (20)
secutor	1 / (1)	..	..	1 / (1)
taeniopus	1 / (1)	..	..	1 / (1)
Deinocerites cancer	..	28 / (1)	61 / (1)	89 / (2)
Psorophora confinnis	..	1 / (1)	..	1 / (1)
Uranotaenia lowii	1 / (1)	..	..	1 / (1)
Total mosquitoes	947	67	131	1145
Total pools	(28)	7	6	(41)
Traps	16	4	4	24
AV/Trap	59	16	32	47

* Number of mosquitoes tested.
** Number of pools tested.
*** Virus isolated from 2 of 18 pools of *Culex nigripalpus*. The first pool consisted of 1 gravid and 49 unfed individuals, and the second, 1 engorged (black blood), 1 gravid and 48 unfed mosquitoes.

TABLE 2.—Complement-fixation identification of J3-16h and J3-16n mosquito isolates.

	Hyperimmune mouse sera										
								Dengue			
Antigen*	EEE	WEE	SLE	ILH	YF	Calif.	Ten	I	II	III	IV
J3-16h	0/64**	0/64	≥64/128	0/128	0/128	0/32	0/128	0/32	0/64	0/16	0/64
J3-16n	0/64	0/64	≥64/128	0/128	0/128	0/32	0/128	0/32	0/64	0/16	0/64

* Crude borate saline antigen.

** Reciprocal of the serum titer with test antigen/reciprocal of serum titer with homologous antigen.

while failing to significantly neutralize Ilheus and Dengue II viruses. Also, rabbit antisera prepared with known strains of SLE virus neutralized the two isolates, while Ilheus and Dengue antisera did not neutralize them.

The recovery of two SLE strains of virus from *C. nigripalpus* collected in August of 1963 represents the second consecutive year the virus was shown to be active in Jamaica. In the absence of a recognized epidemic, these isolations in consecutive years suggest that SLE is endemic on the island.

SUMMARY. Two strains of St. Louis encephalitis virus were isolated from *Culex nigripalpus* in Jamaica in August, 1963. These represent the second and third isolations of SLE virus from mosquitoes in Jamaica.

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Literature Cited

BELLE, E. A., GRANT, L. S., and PAGE, W. A. 1964. The isolation of St. Louis encephalitis

TABLE 3.—Neutralization identification of J3-16h and J3-16n isolates.

	Ascites fluids and sera used							
	Mouse hyperimmune			Rabbit hyperimmune sera				
Virus	J3-16h PR-40446 Ascitic fluid	J3-16n PR-40445 Sera	J3-16n PR-40445 Ascitic fluid	SLE-TBH-28 PR-28916	SLE Parton PR-13759	Ilheus PR-28926	Dengue II PR-30581	Normal control titer
J3-16h (Jamaica)	3.0*	2.5	2.9	4.2	3.9	0.9	<1.0	9.5
J3-16n (Jamaica)	1.9	2.9	2.4	4.0	3.4	0.7	<1.4	8.9
SLE-TBH-28 (Florida)	2.2	2.2	..	3.4	3.4	..	..	8.7
SLE-Parton	1.7	2.0	..	2.8	2.4	..	..	7.6
Ilheus 331 (Brazil)	0.9	..	0.0	..	..	2.0	..	7.9
Dengue II (Tr-1751) (Trinidad)	1.1	..	1.1	..	..	..	2.9	7.1

* $Logs_{10}$ of virus neutralized, measured by intracerebral inoculation of weanling mice.

virus from mosquitoes *(Culex nigripalpus)* in Jamaica. Am. J. Trop. Med. & Hyg. 13:452–454.

CHAMBERLAIN, R. W., SUDIA, W. D., COLEMAN, P. H., and BEADLE, L. D. 1964. Vector studies in the St. Louis encephalitis epidemic, Tampa Bay area, Florida, 1962. Am. J. Trop. Med. & Hyg. 13:456–461.

DOW, R. P., COLEMAN, P. H., MEADOWS, K., and WORK, T. H. 1964. Isolation of St. Louis encephalitis virus from mosquitoes in the Tampa Bay area of Florida during the epidemic of 1962. Am. J. Trop. Med. & Hyg. 13:462–468.

HAMMON, W. McD., and WORK, T. H. 1964. In diagnostic procedures for virus and rickettsial diseases, E. H. Lenette, Ed. (American Public Health Assoc., New York).

REED, L. J., and MUENCH, H. 1938. A simple method of estimating fifty percent end points. Am. J. Hyg. 27:493–497.

SUDIA, W. D., and CHAMBERLAIN, R. W. 1962. Battery-operated light trap, an improved model. Mosquito News 22:126–129.

SUDIA, W. D., and CHAMBERLAIN, R. W. 1964. Experimental infection of *Culex nigripalpus* Theobold with the virus of St. Louis encephalitis. Am. J. Trop. Med. & Hyg. 13:469–471.