

VECTOR COMPETENCE OF A HOUSTON, TEXAS STRAIN OF *Aedes albopictus* FOR RIFT VALLEY FEVER VIRUS

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The recent introduction of *Aedes albopictus* (Skuse) into the Americas has led to concern that this mosquito might serve as a vector of both native as well as exotic viruses (Bartnett 1986, Shroyer 1986, Sprenger and Wuithiranyagool 1986). Experimental and field data, recently reviewed by Shroyer (1986), indicate that this species is capable of transmitting chikungunya, Japanese encephalitis, yellow fever, West Nile, Ross River, and all 4 serotypes of dengue viruses, as well as St. Louis encephalitis and western equine encephalitis viruses.

We evaluated the potential of the F₃ generation of a Houston, Texas strain of *Ae. albopictus* to transmit Rift Valley fever (RVF) virus. This strain of *Ae. albopictus* was provided by Dr. C. Mitchell and is the same strain used in other vector competence studies (Mitchell et al. 1987). The ZH501 strain of RVF virus, isolated from a human case during the outbreak in Egypt in 1977 (Meegan 1979) and passed twice in fetal rhesus lung cells, was used throughout these studies.

Female mosquitoes, 5 to 10 days old reared at 26°C with a 16:8 L:D photoperiod, were allowed to feed on an anesthetized hamster that had been inoculated with RVF virus 24 hours previously. Immediately after the infectious bloodmeal, a sample of the engorged mosquitoes was titrated to determine the titer of virus in the blood they had ingested. The remaining engorged mosquitoes were placed in a 3.8 liter cardboard container with netting over one end. This container was placed in an incubator

maintained at 26°C, and apples or a 5% sucrose solution were provided as a carbohydrate source. The mosquitoes were allowed to oviposit 3 days later, and mosquitoes were assayed for viral infection and transmission ability beginning 7 days after the infectious bloodmeal. During each transmission trial, mosquitoes were allowed to feed either individually, or in groups of up to 5 mosquitoes each on anesthetized hamsters.

After the feeding period, the mosquitoes were cold-anesthetized and their legs and bodies triturated separately in 1 ml of diluent (10% fetal bovine serum in Medium 199 with Hanks' salts and antibiotics). Mosquito or leg suspensions were frozen at -70°C until they were tested for virus by a standard plaque assay on Vero cells (Gargan et al. 1983). Recovery of virus from the body, but not the legs, indicated that viral infection was limited to the midgut and had not disseminated to the hemocoel, while recovery of virus from both legs and body indicated that the mosquito had a disseminated infection (Turell et al. 1984). Hamster death was used as the criterion for virus transmission, because infection with RVF virus is virtually 100% fatal for hamsters (Gargan et al. 1983). All hamsters surviving 21 days after being fed on by a mosquito with a disseminated infection were challenged with 10⁴ plaque-forming units (PFU) of the ZH501 stock virus to demonstrate their susceptibility to RVF viral infection. Ten percent liver suspensions were prepared from a sample of the hamsters that died after exposure to mos-

Table 1. Vector competence of *Aedes albopictus* for Rift Valley fever virus after oral exposure.

	Titer of infectious bloodmeal ¹		
	4.3	4.7	5.9
Infection rate ²	3/90 (3%)	62/124 (50%)	54/61 (89%)
Dissemination rate ³	0/90 (0%)	40/124 (32%)	30/61 (49%)
Transmission rate ⁴	0/85 (0%)	4/110 (4%)	6/49 (12%)

¹ Log₁₀ PFU ingested per mosquito.

² No. infected/no. tested (% infected)

³ Percentage of all mosquitoes (including uninfected) with virus in their legs. Mosquitoes examined 7 to 56 days after the infectious bloodmeal.

⁴ Percentage of refeeding mosquitoes (including uninfected) that transmitted virus. Mosquitoes examined 7 to 56 days after the infectious bloodmeal. Transmissions occurred on days 14 through 56.

quitoes, and evidence of RVF viral antigen was confirmed in each of these suspensions by an enzyme-linked immunosorbent assay (Turell et al. 1986).

Viral infection, dissemination, and transmission rates in *Ae. albopictus* were dose dependent (Table 1). While both infection and dissemination rates were high after ingestion of $10^{5.9}$ PFU of virus (89 and 49%, respectively), transmission was relatively inefficient [6/58 (10%)] for all those refeeding. If only mosquitoes with a disseminated infection were included, 6/28 (21%) transmitted virus. Viral transmission was even less efficient when $10^{4.7}$ PFU were ingested; 4/116 (3%) of all refeeding mosquitoes transmitted virus, including 4/38 (11%) of those with a disseminated infection. Transmission was first observed on day 14 after the infectious blood meal, but earlier transmission might have been observed if larger sample sizes had been used.

In a companion study, *Ae. albopictus* were intrathoracically inoculated with $10^{3.1}$ PFU of RVF virus. Evidence of infection was detected in all mosquitoes tested, with virus titers often reaching 10^6 PFU per mosquito by 4 days after inoculation. These titers were similar to those observed in mosquitoes with a disseminated infection that had been infected orally. There was no significant association between the transmission rate and the number of days between inoculation and the transmission attempt (Table 2). Overall, 13/88 (15%) of the inoculated mosquitoes transmitted virus to hamsters. Similarly, 10/66 (15%) of the orally infected mosquitoes with a disseminated infection transmitted virus.

While the 15% transmission rate for mosquitoes with a disseminated infection appears to be low, a similar rate has been observed for *Aedes mcintoshi* Huang (Turell, unpublished data). This mosquito, [reported as *Aedes lineatopenis* (Ludlow)], has been implicated as one of the

vectors of RVF virus in Africa (Linthicum et al. 1985, McIntosh et al. 1980). The RVF viremias to which mosquitoes were exposed in the present studies are comparable to those observed in young sheep (Easterday 1965, Easterday et al. 1962) and in humans during the outbreak in Egypt (Meegan 1979). Also, *Ae. albopictus* feeds readily on humans, as evidenced by its role in the transmission of dengue virus. Thus, *Ae. albopictus* should be considered a potential vector of RVF virus, should it be introduced into the southern United States.

We thank Dr. Carl Mitchell for providing the *Ae. albopictus* used in this study. In conducting the research described in this report, the investigators adhered to the "Guide for the Care and Use of Laboratory Animals," as promulgated by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council. The facilities are fully accredited by the American Association for Accreditation of Laboratory Animal Care. The views of the authors do not purport to reflect the positions of the Department of the Army or the Department of Defense.

REFERENCES CITED

- Bartnett, R. E. 1986. The perspective of *Aedes albopictus* from the administrative viewpoint. *J. Am. Mosq. Control Assoc.* 2:418-419.
- Easterday, B. C. 1965. Rift Valley fever. *Adv. Vet. Sci. Comp. Med.* 10:65-127.
- Easterday, B. C., L. C. Murphy and D. G. Bennett. 1962. Experimental Rift Valley fever in lambs and sheep. *Am. J. Vet. Res.* 23:1231-1240.
- Gargan, T. P., II, C. L. Bailey, G. A. Higbee, A. Gad and S. El Said. 1983. The effect of laboratory colonization on the vector pathogen interactions of Egyptian *Culex pipiens* and Rift Valley fever virus. *Am. J. Trop. Med. Hyg.* 32:1154-1163.
- Linthicum, K. J., F. G. Davies, A. Kairo and C. L. Bailey. 1985. Rift Valley fever virus (family Bunyaviridae, genus *Phlebovirus*). Isolations from Diptera collected during an inter-epizootic period in Kenya. *J. Hyg. Camb.* 95:197-209.
- McIntosh, B. M., P. G. Jupp, I. Dos Santos and B. J. H. Barnard. 1980. Vector studies on Rift Valley fever virus in South Africa. *S. Afr. Med. J.* 58:127-132.
- Meegan, J. M. 1979. The Rift Valley fever epizootic in Egypt 1977-78. 1. Description of the epizootic and virological studies. *Trans. R. Soc. Trop. Med. Hyg.* 73:618-623.
- Mitchell, C. A., B. R. Miller and D. J. Gubler. 1987. Vector competence of *Aedes albopictus* from Houston, Texas, for dengue serotypes 1 to 4, yellow fever and Ross River viruses. *J. Am. Mosq. Control Assoc.* 3:460-465.

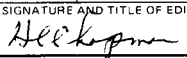
Table 2. Transmission of Rift Valley fever virus by *Aedes albopictus* after intrathoracic inoculation.

Days after inoculation	Transmission rate ¹
4	1/12 (8%)
7	4/15 (27%)
14	3/19 (16%)
21	3/17 (18%)
29	0/6 (0%)
39	1/5 (20%)
48	1/14 (7%)
Totals	13/88 (15%)

¹ No. transmitting/no. feeding (percent transmitting).

Shroyer, D. A. 1986. *Aedes albopictus* and arboviruses: A concise review of the literature. *J. Am. Mosq. Control Assoc.* 2:424-428.
 Sprenger, D. and T. Wuithiranyagool. 1986. The discovery and distribution of *Aedes albopictus* in Harris County, Texas. *J. Am. Mosq. Control Assoc.* 2:217-218.
 Turell, M. J., T. P. Gargan, II and C. L. Bailey. 1984.

Replication and dissemination of Rift Valley fever virus in *Culex pipiens*. *Am. J. Trop. Med. Hyg.* 33:176-181.
 Turell, M. J., C. A. Rossi, R. F. Tammariello, Jr. and C. L. Bailey. 1986. Reduced recovery of Rift Valley fever virus associated with assay of mosquito (Diptera: Culicidae) larval pools. *J. Med. Entomol.* 23:416-422.

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