
A Preliminary Report on Isolations of Arboviruses From Mosquitoes and Migratory Birds¹

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The principle arboviruses that cause human encephalitis in the United States are Western equine encephalitis (WEE), St. Louis encephalitis (SLE), and Eastern equine encephalitis (EEE). Birds serve as natural hosts for these three arboviruses and mosquitoes of the genera *Culex* and *Culiseta* have been involved. Human beings and horses are thought merely to be dead-end hosts (Hess and Holden, 1958).

This paper describes a study of the natural history of certain arboviruses in long range migratory shore birds and mosquito populations. The ecological distribution of these viruses limited this study to WEE and SLE viruses.

MATERIALS AND METHODS

Mosquito and migratory bird blood collection sites were established at the Kansas Wildlife Refuge, Great Bend, Kansas and 30 miles south at Quivera National Wildlife Refuge by the field team (Dr. David Parmelee, Kansas State Teachers College, Emporia, Kansas, and Mr. H. Stephens).

The mosquito specimens were collected during October, 1963, and from April to October, 1964. They were classified according to Carpenter and LaCasse (1955) and pooled in groups of approximately 50. The pools were homogenized in phosphate buffered saline plus 20% calf serum and Penicillin and Streptomycin (PBS) and were centrifuged. A litter of 6 suckling mice was inoculated with 0.05 ml of the supernatant fluid intraperitoneally and 0.03 ml intracerebrally. The mice were observed for a period of 10 days. Brain material from mice which died after the initial 24 hours was studied to detect the presence of viral agents.

¹This work was supported by grant number AI 05232-02, from the National Institutes of Allergy and Infectious Disease, U. S. Public Health Service.

The shore birds were bled by venipuncture of the juglar vein. PBS was added to the bird blood to make a total volume of 1 ml. In order to determine the viral content, the frozen, clotted blood was homogenized in a tissue grinder, appropriate dilutions were made with PBS and were injected into suckling mice as described above. Part of each specimen was retained for studies on antibody content.

RESULTS

There was a total of 16,460 mosquitoes collected and classified. The following numbers of species of mosquitoes were classified: *Culex tarsalis*, 4,470; *Culex pipiens*, 298; *Culiseta inornata*, 342; *Aedes dorsalis*, 6,295; *Aedes vexans*, 863; *Aedes nigromaculus*, 1,184; *Aedes sollicitans*, 763; *Aedes* spp. (not classified), 2,205; *Anopheles punctipennis*, 2; *Psorophora cyaneescens*, 2; *Psorophora ciliata*, 3; *Psorophora* spp. (not classified), 32; *Orthopodomyia signifera*, 1.

From 86 pools of *Culex tarsalis*, 22 agents, lethal to suckling mice, have been isolated. One has been identified as Western Equine Encephalitis. From 8 pools of *Culex pipiens*, 5 agents which kill suckling mice have been isolated. No agents were isolated from 10 pools of *Culiseta inornata*.

Twenty of a total of 131 shorebird blood specimens killed suckling mice and will be involved in further identification studies.

DISCUSSION

The collection dates of the mosquitoes which have been implicated as containing viral agents correlate well with the facts that WEE occurs in midsummer and SLE in late summer or fall. The suspected mosquito pools all were collected from July to October. WEE antibody appears in chickens during late August or September in the midwest, therefore, infected mosquitoes would be present shortly before this time.

Upsetting the natural cycle of the mosquito-bird relationship may cause the involvement of man. Increased mosquito prevalence and/or decreased avian host population may lead to human infection. Control of birds that serve as epidemic hosts of the viruses may lessen epidemics (Hess and Holden, 1958). Long term sentinel studies may be used to predict outbreaks so that preventive measures can be taken (Sudia and Chamberlain, 1959).

SUMMARY

From 16,460 mosquitoes collected and classified, 104 pools were tested for viral content and 27 agents lethal to suckling mice are being identified. One virus has been identified as WEE. One hundred and thirty-one long range migratory bird blood specimens have been studied for viral content, and 20 have been shown to contain agents lethal to young mice. They are being identified.

We wish to thank John Janovy, Jr. for assistance in the classification of mosquitoes.

LITERATURE CITED

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