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Authors: HOFF, GERALD L., and BIGLER, WILLIAM J.

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HUMAN ENTEROVIRUSES AND WILDLIFE: ISOLATION FROM GRAY SQUIRRELS

GERALD L. HOFF[□] and WILLIAM J. BIGLER, Health Program Office, Florida Department of Health and Rehabilitative Service, 1317 Winewood Blvd., Tallahassee, Florida, 32301, USA.

FLORA MAE WELLINGS and ARTHUR L. LEWIS, Laboratory Services, Florida Department of Health and Rehabilitative Services, 4000 Buffalo, Tampa, Florida, 33614, USA.

Abstract: Five isolates of human echovirus 1/8 complex were recovered from the feces of free-ranging gray squirrels. The source of infection and the significance of the isolates remain unknown.

INTRODUCTION

A number of reports concern the isolation of human enteroviruses from animals and these have been summarized by Graves and Oppenheimer.¹ Polio, coxsackie and echoviruses all have been isolated; however, the coxsackie viruses seem to predominate. Of the 15 species of mammals and birds listed from which these viruses have been recovered, only two, house crows (*Corvus splendens*) and monkeys (*Presbytis entellus*) were not domestic or peridomestic species.⁴ Coprophagy and drinking of contaminated surface waters were believed to be the mechanism of virus exposure.

Since a diverse array of wildlife species inhabit the urbanized areas of Florida, the State Department of Health and Rehabilitative Services has focused considerable attention upon urban wildlife as hosts of zoonotic diseases and monitors of environmental quality.^{1,2} Therefore, as part of a one year multifaceted investigation on the gray squirrel (*Sciurus carolinensis*) population in the city of Jacksonville, the occurrence of human enteroviruses in this species was studied.

MATERIALS AND METHODS

Jacksonville is located in northeast Florida. The city covers 2200 km² and is

intersected by two major rivers and numerous streams. At the time of the study, there were approximately 50 major raw sewage outfalls into the waterways.

The trapping design and methods, and techniques of age determination have been described previously.⁶ Squirrels were killed by exsanguination, and the fecal pellets recovered by either stripping the lower intestinal contents or collecting freshly excreted materials. The pellets were kept at -70 C until assayed for the presence of virus. Standard enterovirus examination procedures were followed.⁸

RESULTS

A total of 180 gray squirrels were captured from 36 sites (5 squirrels/sites; 3 sites/month). The squirrels ranged in age from <1 year to 4 years old, with 80% of the animals <2 years old.

Five viral isolates from 5 different squirrels were recovered in African green monkey kidney cell culture. Through pooled immune sera and neutralization tests with specific immune sera all the isolates were identical and were identified as members of the Echovirus 1/8 complex.⁷ Three of the squirrels which yielded virus were captured at the Jacksonville Zoo, while the other 2 squirrels were from two different residen-

[□] Current Address: Kansas City Health Department, 1423 Linwood Blvd., Kansas City, Missouri 64109, USA.

tial areas of the city. Four of the isolates were recovered in January and the fifth in October, 1974. Two of the squirrels were < 1 year old, two were one year old, and one was 2 years old.

DISCUSSION

While these isolations represent the first reported recovery of human enteroviruses from terrestrial wildlife in Florida, their significance remains unknown. Questions remaining unanswered are 1) do the members of the ECHO 1/8 complex cause disease in the squirrels; is it part of the squirrel's normal intestinal flora; or are they transients; 2) do the viruses come from contact with human fecal wastes or some other source; and 3) does the squirrel act as transmitter of the ECHO 1/8 complex to man or his pets.

Human enteroviruses may be carried for several months in the gastrointestinal tracts of dogs, but are not part of the normal flora and are derived from an unknown external source.³ Dogs do not show clinical signs unless fed large doses

of virus.⁹ A preliminary attempt to infect squirrels *per os* with the ECHO 1/8 complex virus isolated, failed to establish detectable enteric infection. Also, based on sero-epidemiology, antigenic characteristics, and RNA hybridization studies, it has been postulated that swine vesicular disease virus is recently derived from human coxsackie B5 virus.⁵ A similar situation may exist with human ECHO 1/8 complex viruses and the isolates from the squirrels.

The source of virus for the squirrels remains undetermined although given the waste disposal situation in the city, it is easy to speculate that contaminated surface waters were involved. Four of the infected squirrels were captured within 0.11 km of the rivers while the fifth squirrel was captured 2.5 km from any surface water. Throughout the study, squirrels were observed entering the sanitary-storm sewer system, presumably seeking water. Regrettably, this possible source of infection remains speculation, and since the study all raw sewage outfalls have been eliminated by the city.

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