ELECTRON MICROGRAPHS OF NEWCASTLE DISEASE VIRUS PROPAGATED IN THE CAVE BAT (MYOTUS LUCIFUGUS)

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Electron microscope studies were conducted to determine the morphology of the Newcastle disease virus after intracerebral passage in Myotus lucifugus

Figure 1. Newcastle disease virus from the brain of an infected cave bat, Myotus lucifugus (lsc), after one subculture in embryonated eggs. $\times$ 122,500.

the cave bat. Bang (1947, 1948) and Cunha et al. (1947) showed a predominance of tailed virus forms from purified egg-adapted virus material after the virus had been suspended in 0.07 to 0.15 molar sodium chloride. These tailed forms
were also found in hamster-adapted, mouse-adapted, and monkey-adapted Newcastle disease virus after culture in 10-day embryonated eggs by Reagan and his co-workers (1949, 1950).

Five-hundredths ml of allantoic fluid from 10-day embryonated eggs infected with the twenty-fourth egg passage of California strain (no. 11,914) was in-

![Figure 2. Newcastle disease virus from the brain of an infected cave bat, *Myotus lucifugus* (lsc), after one subculture in embryonated eggs. × 90,000.](http://jb.asm.org/)

oculated into the right frontal lobe of each of 4 *Myotus lucifugus* cave bats. Within 4 to 6 days, the bats showed typical Newcastle disease symptoms: irritability and malaise followed by rhythmical contraction of muscles starting in the limbs and spreading over the entire body, paralysis of the hind legs, prostration, and death. There was no evidence of excessive salivation from pharyngeal paralysis, which is characteristic in other animals infected with the hamster-
adapted Newcastle disease virus. When nervous symptoms developed in the bats, the brain was removed aseptically, ground with alundum in a small mortar, and diluted to a 10 per cent suspension with physiological saline. This virus-bearing suspension was then injected intracerebrally into each of 6 normal bats as described above. All bats were kept separately in small animal jars and fed daily with food containing ground banana, meal worms, cottage cheese, egg yolk, wheat germ oil, and multivitamins.

When bats of the first and second passage showed typical Newcastle disease symptoms from 3 to 6 days after intracerebral inoculation, they were sacrificed, and the same procedure was carried out in making a 10 per cent brain suspension. This virus-bearing material was inoculated into the allantoic sac of twelve 10-day embryonated chicken eggs. All embryos died within 72 hours. A pool of the infected allantoic fluid from the dead eggs was prepared. The presence
of virus in this material was demonstrated and the virus was verified as that of Newcastle disease by a neutralization test using embryonated chicken eggs as the test host.

Allantoic fluid from both the first and the second bat-passaged egg material was centrifuged in a horizontal centrifuge for 5 minutes at 1,000 rpm. The sediment was discarded, and the supernatant was recentrifuged for 1 hour at 44,620 rpm in a Spinco ultracentrifuge. There was less than a 1-degree change in the temperature of the refrigerated outer jacket. The sediment was resuspended with 3 per cent saline for 5 minutes, placed on parlodion-prepared screens, shadowed with chromium (Williams and Wyckoff, 1945a, b, 1946) at arc tan 1/10 and examined under the RCA EMU electron microscope.

Various fields chosen for electron examination showed many virus particles with uniform tail structure. Figures 1 and 2 show typical tail forms from the first bat passage after virus cultivation in embryonated eggs. These tails appear to be segmented. The high magnification brings out great detail in these virus studies. Figure 3 shows typical tail forms from the second bat passage after cultivation in embryonated eggs. These tails appear to be segmented, as shown also in figure 1.

**SUMMARY**

Bat-passaged Newcastle disease virus from the first and second subinoculations in bats by the intracerebral route of inoculation was 100 per cent pathogenic for embryonated chicken eggs on the first embryo passage. The virus in the allantoic fluid of eggs inoculated with the bat brain material was neutralized by positive Newcastle disease serum but was not affected by normal chicken serum.

Electron micrograph studies of these virus particles at high magnification seem to bring out more detail in the taillike structure of this virus.

**REFERENCES**


