RESEARCH NOTE

FURTHER OBSERVATIONS ON THE ULTRASTRUCTURE OF EPHEMERAL FEVER VIRUS

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Electron microscopic studies on ephemeral fever virus (EFV) have shown that the virus is cone-shaped having a basal diameter of approximately 176 m μ and a height of about 88 m μ (Lecatsas, Theodoridis & Erasmus, 1969). These workers have also shown that the virus has an envelope derived from the host cell membrane and that the internal electron-dense component consists of a spirally arranged material, probably ribonucleoprotein in nature. The length of the spiral is between 2 and 2.5 μ with a pitch of about 16.5 m μ .

Further electron microscopic observations confirm previous results and provide new information on the ultrastructural nature of the electron-dense spiral. These are reported in the present communication.

The methods of preparation of material have been reported previously (Lecatsas *et al.*, 1969) and consist basically of fixation of virus-infected BHK 21 cells with gluteraldehyde followed by osmium tetroxide, dehydration in ethanol and embedding in Epon 812. Thin sections were then double-stained with uranyl acetate and lead citrate and examined in Siemens Elmiskop 1A electron miscroscope.

Close examination of the arrowed portion of the virus particles in Plate 1 (1) reveals that the granular electrondense material represents sectioned strands. It is also evident that the electron-dense material is helical in nature (arrows). Measurement of the individual strands gives an approximate thickness of 4 m μ (40 Å). An enclosed space between the strands is clearly visible in Plate 1 (3). The total diameter of limb "a" of the virus particle shown in Plate 1 (3) is 18.4 m μ , and that of limb "a" in Plate 1 (4) is 16.8 m μ . The sharp margin of both limbs suggests that the virus particles have been cut by a plane passing through the core axis. It is also evident that the helical nature of the strands and the space they enclose would give a value near 17 m μ if two strands of 4 m μ each make up the helix. Allowing for the relatively large space (8 m μ) between the strands which is evident in Plate 1 (4), three or more strands would yield a larger total diameter. Plate 1 (3 and 4) suggests strongly that the strands form a double helix. The axial length of one complete turn of the helix is about 10 m μ (100 Å). Plate 1 (2) is a proposed model of the virus.

It is interesting to note that Dunnebacke & Kleinschmidt (1967) have shown that the total length of reovirus ribonucleic acid (RNA) is about 5 m μ . The observations made in the present investigation suggest that the length of RNA in EFV lies between 4.5 and 6 m μ .

It remains to be seen whether the RNA contained by each 4 m μ strand is single or double stranded. Biochemical characterization of purified EFV RNA should provide further insight into the arrangement of the hereditary material in this unique virus.

References

- DUNNEBACKE, T. H. & KLEINSCHMIDT, A K., 1967. Ribonucleic acid from reovirus as seen in protein monolayers by electron microscopy. Z. Naturf. 22b, 159-164.
- LECATSAS, G., THEODORIDIS, A. & ERASMUS, B. J., 1969. Electron microscopic studies on bovine ephemeral fever virus. Arch. ges. Virusforsch., 28, 390–398.

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- PLATE 1 (1) Virus particles in the cytoplasmic matrix. Arrows indicate areas where helical nature of electron-dense material is evident. Bar equals 80 m μ .
- PLATE 1 (2) Proposed model of EFV. The space separating the individual strands is not shown in the model.
- PLATE 1 (3) Helical array of strands composing the electron-dense portion of EFV is indicated by the rectangle. Diameter of the limb "a" is 18.4 mµ. Bar equals 80 mµ.
- PLATE 1 (4) Demarcated portion of virus particle suggesting helical array of electron-dense strands. Space between strands is evident. Diameter of the limb "a" is 16.8 m μ . Bar equals 60 m μ .