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Preliminary Report

Micromorphological Changes in Measles Infected KB Cells

There have been few reports on the intracellular development of measles virus though the tracing of the behavior of virus particles in the cell is very important for the full understanding of the nature of measles virus.

Kallman *et al.*[2] and Tawara *et al.*[3] described the micromorphology of the intranuclear and cytoplasmic inclusions in measles infected HeLa cells and Baker *et al.*[1] reported the existence of crystallites in the nucleus of measles infected human amnion cells.

In the course of a morphological study of the development stages of measles virus in KB cells the authors observed two new sturctures associated with virus infection, one in the peripheral region of the nucleus and the other in the cytoplasm.

KB cells were grown in YLH medium (containing 20% bovine serum) for seven days and on the seventh day KB adapted Toyoshima strain[4] virus was inoculated. Samples for electron microscopy were taken five days after this inoculation. At this stage of infection there was a marked cytopathic effect in many of the cells in the culture.

The infected cells were detadched from the bottle wall by gently shaking the bottle and were centrifuged for five minutes at 1,000 rpm.. After fixation in 1 per cent osmium tetroxide in Veronal-acetate buffer, pH 7.4, for 1 hour at 4°C, the pellets were dehydrated through the ethanol series (25, 50, 75, 95 and 100 %) and embedded in Epon 812 according to Luft's method. Sections (ca. 250 Å thick) were stained with 1 per cent uranyl acetate in 75 per cent ethanol and examined in a Hitachi HU-9D electron microscope.

Fig. 1 shows our first new observation, namely the vesicular structure in the nucleus. Spherical electron-dense particles are observed inside the vesicle. The diameter of the particles is 100-150mμ and some of them have a central dense mass and outer limiting membrane. The shape of the vesicle is not regular. It is always in the periphery of the nucleus.

Our second new observation is the intracytoplasmic vesicles, illustrated in Fig. 2. Electron-dense particles of 40mμ diameter and short filaments of 30-40mμ width are observed in the vesicle. Furthermore fine particles of 10mμ diameter were found in the vesicle. The membrane of the vesicle is associated with fine electron-dense particles.

Besides these new findings the intranuclear and intracytoplasmic inclusions described by Kallman *et al.* and Tawara *et al.* were also seen.

Our findings are quite different from those described by others and, at present, we have no ideas on the correlation of our findings with those of Kallman *et al.*, Tawara *et al.*, or Baker *et al.*. We are rather inclined to Kallman's opinion[2]

that "the strands seen in the inclusions can not be fully formed virus".

It may be suggestive to indicate that the particles in intranuclear vesicles are almost the same size as the measles virus particles recently observed by Waterson *et al.*[6] by the negative staining method. It is interesting to consider the relationships between our findings and the compact measles virus antigen accumulating in the paranuclear areas of infected FL cells observed by Toyoshima *et al.*[5] by the immunofluorescence technique.

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EXPLANATION OF PHOTOGRAPHS

Fig. 1. An electron micrograph of an intranuclear vesicle found in measles infected KB cells. Eletron-dense particles of 100-150 mμ in diameter are seen inside the vesicle. N : nucleus Magnification: ×59,800.

Fig. 2. An electron micrograph of an intracytoplasmic vesicle in measles infected KB cells. Several forms of electron-dense particles can be seen in the vesicle. Magnification: ×59,800.

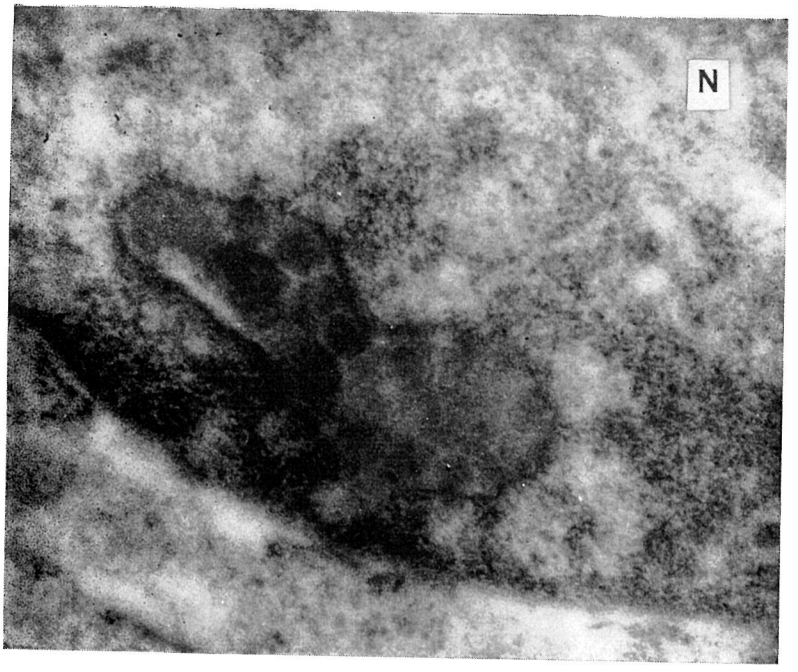

Fig. 1.

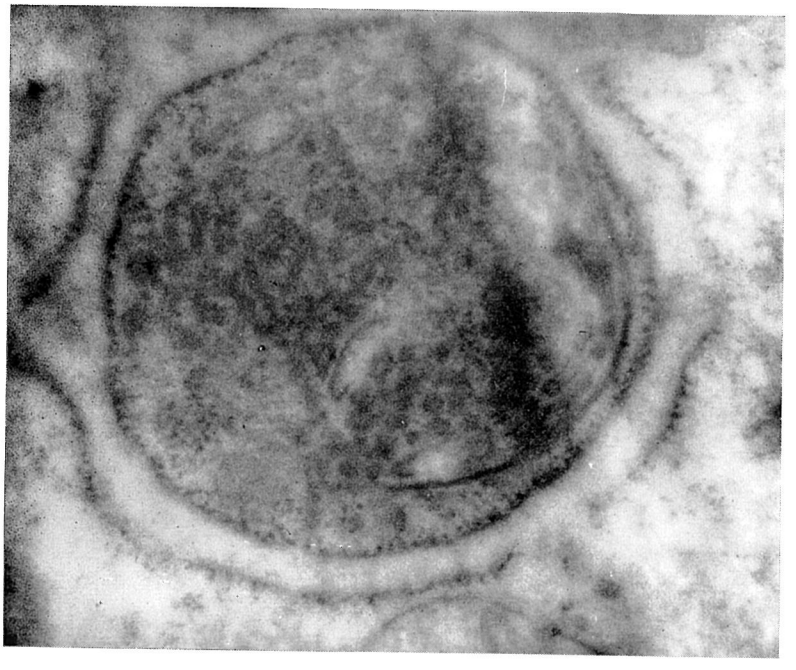

Fig. 2.