GLOBOID STRUCTURES IN THE CYTOPLASM OF RAPIDLY GROWING HELA CELLS*

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We have used electron microscopy of sectioned cells in order to study a group of viruses associated with acute respiratory infections in human beings and have found that virus-like particles are located within the nuclei of infected cells (1, 2). The HeLa cells used for this group of viruses were cultivated originally from a human cervical carcinoma (3) and have been transmitted serially in many laboratories for use in viral investigations. The present observations were made during studies of uninfected HeLa cells carried out in order to look for virus-like particles unrelated to the group under study and also to learn about characteristics of the host cell that might have bearing on susceptibility or resistance to infection.

Methods

Tissue culture of HeLa cells was carried out essentially as described by others (4, 5). Rapid growth of cells took place in 1 to 2 days in a medium consisting of equal parts of human serum, tryptose phosphate (6), and balanced salt solution.[1] Sectioning techniques and electron microscopy were as used before (7) with the following exceptions: In addition to the fixative of Palade (8), we have used a similar fixative containing dichromate described by Dalton (9). Duration of fixation varied from 30 seconds to 30 minutes. Cells were allowed to remain *in situ* on the glass tube during fixation and subsequent application of solutions; they were removed from the glass just before embedding.

RESULTS

In rapidly growing cultures of uninfected HeLa cells, it was found that the cytoplasm contained large numbers of unusual bodies. These structures were round or ovoid in shape, somewhat larger than mitochondria, and were surrounded by membranes that were almost always single. The internal structure of the bodies showed considerable variation, but large amounts of electron-dense material were usually present (Figs. 1 and 2). Small round or ovoid struc-

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[1] The composition of the balanced salt solution is given in reference 4.

tures were often associated with the electron-dense material as if embedded within it. Vacuoles surrounded by membranes and other electron-lucent areas were also present within the bodies. Cristae typical of mitochondria were not demonstrated definitely, but some structures were seen that might be interpreted as transition forms. The bodies were distinguished readily from the dense irregular cytoplasmic granules previously described (1, 2) by their limiting membranes, internal structure, and relatively lower density. As a test of adequate fixation, it was noted that well preserved examples of the Golgi complex and other structures could be seen in areas of the cytoplasm adjacent to the globoid bodies (Fig. 3).

HeLa cells kept in maintenance solutions at 30° or 36°C. grew slowly or not at all, and under these conditions, the globoid bodies were few in number or absent. The few globoid bodies in such relatively resting cells were found to have less electron-dense material, and the internal structure was more prominent (Fig. 4).

DISCUSSION

Globoid structures in the cytoplasm of uninfected HeLa cells have interest because they occur in large numbers under conditions of rapid growth and because HeLa cells were derived from a carcinoma. The following points suggest that they are mitochondria: (*a*) similar structures containing vacuoles, membranes, and electron-dense material have been described in other types of cells and are thought to be mitochondria (10, 11); (*b*) some mitochondria contain microvillus-like structures appearing in sections as circles (12–14); (*c*) electron-dense material accumulates in damaged mitochondria (11, 12, 15); and (*d*) phase cinematography of cells in tissue culture has shown that mitochondria may assume globoid shapes and contain vacuoles (16).

SUMMARY

During the course of electron microscopic study of rapidly growing uninfected HeLa cells, it was found that numerous globoid bodies occurred in the cytoplasm. Reasons are given for suspecting that these structures are mitochondria.

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EXPLANATION OF PLATE 115

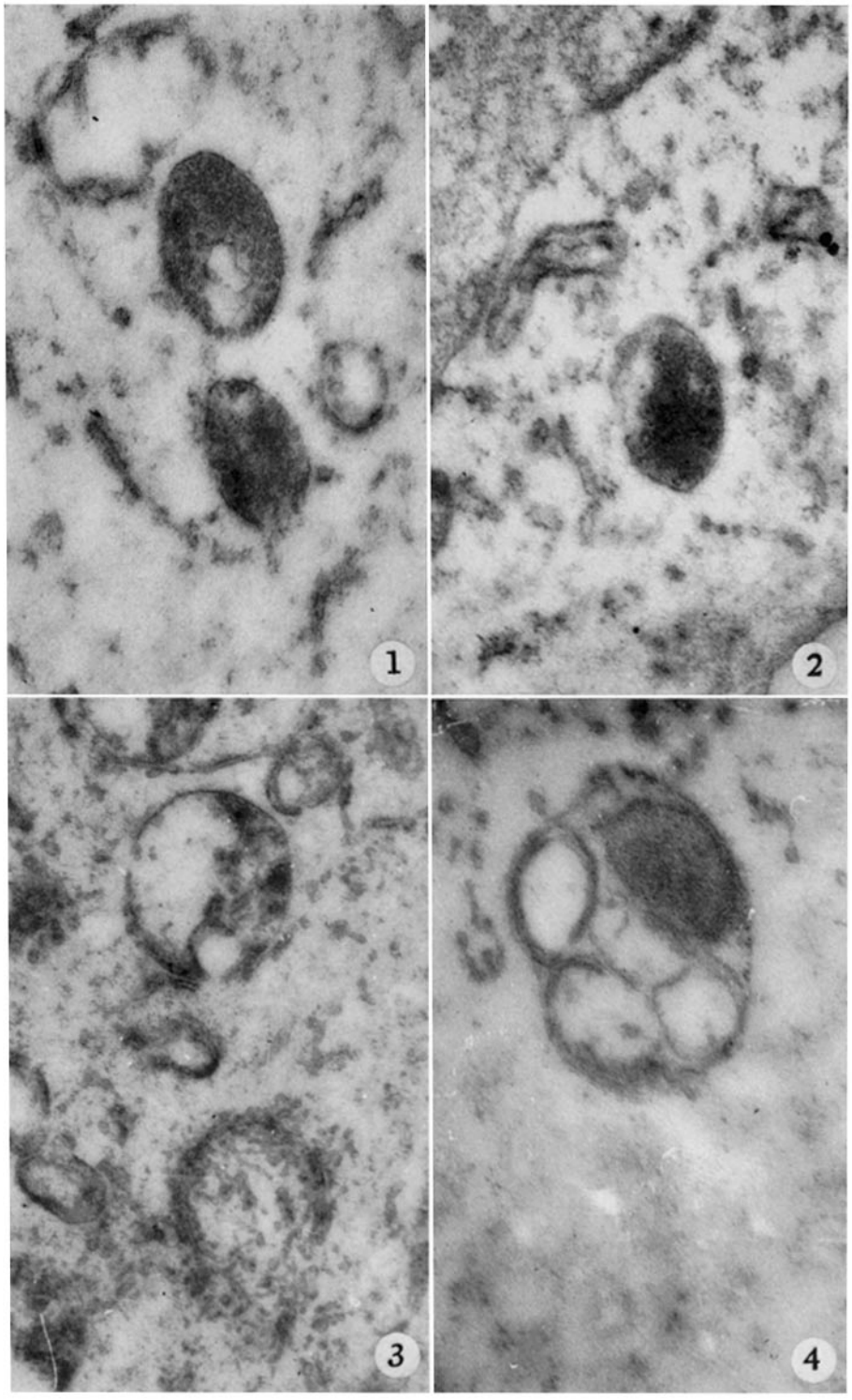
Photographic prints were made by Mr. Cramer Lewis.

FIG. 1. Globoid bodies in the cytoplasm of a rapidly growing HeLa cell 48 hours after transplantation. The upper body contains a vacuole surrounded by a membrane. Both bodies contain electron-dense material and small indistinct circular structures. Both bodies are surrounded by single membranes. Dalton's fixative for 30 seconds. Magnification approximately 30,000.

FIG. 2. Globoid body in rapidly growing cell 24 hours after transplantation. Electron-dense material within the body is associated with small indistinct circular structures. The body is surrounded by a single membrane. The nucleus of the cell appears at the upper left. Dalton's fixative for 30 minutes. Magnification approximately 49,000.

FIG. 3. Cytoplasm of rapidly growing cell 22 hours after transplantation. In the upper part of the field is a body surrounded by a membrane which is single except for a small lower section where it is double. The body contains less electron-dense material than the bodies in Figs. 1 and 2. In the lower part of the field is a well preserved example of the Golgi complex similar in size to the globoid body. Dalton's fixative for 1 minute. Magnification approximately 30,000.

FIG. 4. Globoid body in the cytoplasm of a HeLa cell kept in a maintenance solution of yeast extract and glucose (17) at 30°C. for 3 days. An area of electron density, 4 vacuoles, and the body itself are surrounded by membranes. Dalton's fixative for 1 minute. Magnification approximately 30,000.



(Harford *et al.*: Globoid structures in cytoplasm of HeLa cells)