MORPHOLOGIC STUDY OF VIRUS-LIKE PARTICLES IN A CASE OF ACUTE LEUKEMIA

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Virus-like particles, grouped in clusters not bound by a membrane, were seen in electron micrographs of fresh leucoblasts of an acute leukemic patient. They consisted of large (100-nm diameter), round particles apparently composed of subunits. Tubular structures (30–50-nm diameter) were also seen in leucoblasts of the same patient. The two types of structures were never seen simultaneously in the same cell. The authors review the literature pertaining to the morphology of virus-like particles associated with malignant hematologic disease.

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Although they are sought meticulously with the electron microscope, virus-like particles have been observed rarely in examination of white blood cells in human malignant hematologic diseases.

We compared the virus-like particles described in the literature with oncogenic viruses and schematically classified them into the following four categories.

Epstein-Barr Virus

This is the only virus found regularly in certain types of hematosarcoma; it has been found in cell cultures and even in fresh tumor cells of Burkitt tumors. It has been found in cell cultures of Kaposi sarcoma and rarely in cultures of cells of chronic myeloid leukemia.

Particles Observed in the Plasma of Leukemic Patients

They have been observed mainly by Dmochovski et al. and Levine.

Tubular Structures

These structures found in various diseases in man and under experimental conditions have been recently reviewed by Lyon. Their nature and significance have been the source of much discussion.

Non-Identified Particles

At present, these particles do not fall into any of the above categories. Their viral nature has not yet been proved but their ultrastructure suggests that it might be possible. The shown structures and those described by Tavassoli and Baughan and Seman are in this category.

In the present case, the patient was a 73-year-old woman whose white blood cell count was found to be 270,000, of which 98% were leucoblasts. A diagnosis of acute leukemia was made. The patient died several days after admission.

Light Microscopy of the Blood Smears

In smears stained with May–Grünwald Giemsa large, round leucoblasts were seen; their nucleocytoplasmic ratio was rather elevated. The oval-shaped nucleus was skewed to one side of the cell and limited a rather large, clear cytoplasmic zone. In the nucleus, the chromatin, which was uniform in aspect, was fine and dispersed and there were one or two nucleoli.

The cytoplasm was sparse and only slightly basophilic; it did not contain any granules. At this magnification, it seemed to be rather homogenous (Fig. 1).

Electron Microscopy

In view of the poorly differentiated aspect of the blood smears, the white cells were processed for electron microscopy.
The leucoblasts were round and small (between 8 and 14 μm) and rather monomorphic, with a very large, often kidney-shaped nucleus. Depending on the section, one or two nucleoli were present (Fig. 4). The chromatin was completely dispersed without any margination but the internal nuclear membrane was particularly dense (Fig. 2).

The cytoplasm, more or less abundant, was distributed in fibrillar, very often concentric formations, in most of the cells. These formations were closed and sometimes adjacent to the nucleus and to the numerous mitochondria (Figs. 2 and 3). These mitochondria were round or slightly elongated with more or less distorted cristae. Some of the cells contained rare azurophilic granules (Fig. 3). In addition, the cytoplasm contained some rather particu-
FIG. 3. Detail of the "vortiginous" plaques of fibers (×28,000). These structures invade a large part of the cytoplasm and seem to be closely related to the nuclear membrane and other organelles.

FIG. 4. Cell containing "large" virus-like particles (×19,500). This cell section shows the presence of about 80 particles distributed in clusters which are not bound by a membrane. The reticular endothelium (er) is rather plentiful and dilated.
Virus-Like Particles in Leukemia

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FIG. 5. Detail of clusters of “large” particles (×50,000). They do not have a completely regular aspect. They seem to be composed of subunits.

FIG. 6. Detail of “large particles” (×225,000). There is no doubt about the existence of subunits.

lar particles and tubular structures which were never seen simultaneously.

The larger-sized particles (100 nm in diameter) present in 5% of the leucoblasts were round, though occasionally elongated (Fig. 5). Twenty to 50 particles were grouped in clusters, never bound by a membrane. The content of each particle was not homogenous and at higher magnifications (Fig. 6) seemed to be composed of juxtaposed subunits.

The tubular structures, present in 8% of the leucoblasts, formed clusters measuring 30–50 nm in diameter (Fig. 7). They were seen most of the time very close to the nucleus. Like the large-sized particle clusters, these tubular arrangements were never bound by a membrane. The tubes were twisted and seemed to be entangled with each other. They were found near the endoplasmic reticulum, the diameter of which was much larger than that of the tubules. Sometimes (Fig. 7) these structures seemed to be contiguous to the mitochondria. High magnification (Figs. 8 and 9) did not furnish additional data.

Discussion

Nature of the Leukemia

The leukemia evolved so rapidly that certain classic explorations could not be performed. This is the reason why cytochemical and enzymologic investigations were not carried out. Nevertheless, from the optical and electron microscope images seen, it seems that this case can be classified as an acute, myeloblastic, poorly differentiated leukemia.
FIG. 8. Detail of tubular structures (×50,000). Vacuoles (v) are present in the heart of these clusters.

FIG. 9. High magnification of the tubules (×150,000). The tubules are sectioned in various planes.
Interpretation of the Various Images Seen

Fibrillar plaques: Vertiginous, more or less concentric fibrillar masses have been frequently found in cancer cells\(^1\) and in lymphoid cells of acute lymphoblastic\(^2\) or myeloblastic leukemia,\(^3\) but their significance is not known.

Large particles: In view of their size and general aspect, these particles appear to be like those described by Seman.\(^4\) However, they seem to be somewhat different because they did not have a dense inner core and were not surrounded by a membrane.

Tubular structures: They look very much like the tubular inclusions referred to before,\(^5\) but the clusters were never bound by a membrane as often described in the literature.

The fact that virus-like particles and tubular structures were found in the leucoblasts of the same patient suggests two hypotheses: 1) The tubular structures are required for the manufacture of the virus-like particles, as Oshiro has shown.\(^6\) However, we never observed both types of structures simultaneously in the same cells and the size of the particles we observed did not correspond to those described by Oshiro. 2) The tubular structures are the response of the cell to a particular stimulus which, in our case, may be the virus-like particles.

The structures we describe do not seem very different from certain virus-like particles already seen by some investigators, but it is impossible to be sure that they are identical or that they really represent viruses.

ADDENDUM

While this article was in press, an article by J. C. Cawley and A. Karpas, “The Ultrastructural Demonstration of Virus-like Particles in Human Leukaemic Cells,” was published in European Journal of Cancer 10:559-561, 1975. The particles observed by these authors resembled exactly those we describe. They also report that two forms of virus-like particles were present, which corresponds to our observations. Their conclusions agree with our’s in that they are purely speculative. It must be noted that these particles have now been observed in several types of leukemia.

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