## THE "SPINDLE-SHAPED BODY" IN FIBROBLASTS

HERBERT VOELZ. From the Department of Microbiology, Indiana University School of Medicine, Indianapolis

In a recent study of the fine structure of normal uninfected fibroblasts from regenerating tendon, periarticular granulation tissue (rabbit), and various other tissues (human, chicken, hamster), Movat and Fernando (1) have described a cell organelle not yet found elsewhere. This organelle is spindle-shaped and occurs irregularly within the cytoplasm, mainly in the Golgi region and in the peripheral portion of the cell. The inner structure of the organelle is apparently fibrous, bundles of fibrils being enveloped in a sheath or membrane. Staining of the cells with phosphotungstic acid showed that the fibrils had a high electron opacity, whereas the sheath or membrane possessed a lower affinity for the stain. The so called "spindle-shaped body" measures 500 to 600 A by 2,500 to 3,000 A. The function of these bodies is not known. However, their relation to secretion has been presumed. Another presumption, that these organelles may be phagocytosed collagen, could not be confirmed since no cross-banding has been detected by the staining technique employed.

In our current studies on the reproductive cycle of Rous sarcoma virus (RSV) in chicken fibroblasts, we have only once encountered cell organelles similar to the "spindle-shaped body" described by Movat and Fernando. It is of general interest to compare, in respect to occurrence and fine structure, the "spindle-shaped bodies" described by these authors with the organelles we have found in cultured chicken fibroblasts.

# MATERIALS AND METHODS

Secondary cultures of chicken fibroblasts were grown in Eagle's medium plus 10 per cent horse serum at 37°C. Parallel cultures were infected with RSV, adjusted to 1 particle per cell. Samples were taken

at several time intervals and prepared for electron microscopy.

The cells were removed from the glass wall of the culture flask by treatment with trypsin and washed twice in a balanced salt solution. A pellet obtained by centrifugation of the fibroblasts was fixed in 2 per cent OsO<sub>4</sub> buffered with veronal-acetate pH 7.4, dehydrated in alcohol, and embedded in Epon-Araldite mixture (2). Thin sections were cut and stained with lead (3) and observed under an RCA EMU-3E at 50 kv.

#### **OBSERVATIONS**

In regard to fine structure, number, and distribution within the cytoplasm, size, and electron opacity, it is concluded that the organelles we have found are the same in both fibroblasts infected with RSV and normal fibroblasts when both are grown in the same medium under the same conditions. The organelles in our preparations are more rod-like than spindle-like in shape (Fig. 1). Spindles, as described by Movat and Fernando and seen occasionally in our electron micrographs, may be ascribed to oblique sections through rodshaped fibers. The width of the organelle is consistent with that given by Movat and Fernando; however, the length is not measurable because of the difficulty of obtaining the entire rod in one plane. It is possible that these structures may extend throughout the entire cell. Their location within the cell seems not to depend upon any structural feature of the cell such as the Golgi region. While Movat and Fernando found that the "spindle-shaped bodies" occur singly and somewhat isolated in the cytoplasm, we have observed that they may also occur in bundle-like pattern (Fig. 2). Longitudinal as well as cross-sections through the organelles (Figs. 2 and 3) show

bundles of fibrils loosely enveloped by a sheath or membrane of the same density when stained with lead. One fiber is estimated to consist of as many as 50 fibrils, each measuring about 50 A in diameter. Since no cross-banding could be demonstrated by our technique, the possibility is excluded that these fibers are collagen in nature.

Another interpretation of the nature of these structural elements suggests that they may be formed by the rough-surfaced endoplasmic reticulum, their contents being a phagocytized or secretion product of the cell. In cells which do not form these structures, one can find similar arrangements of the endoplasmic reticulum as compared with the organelles. In thin sections, only portions of a continuous endoplasmic reticulum can usually be seen as it might be in the case of the rod-like structure, the latter being straightened by turgescence. Further support for this suggestion may be found in an oblique section through a portion of an organelle (Fig. 1, arrow). It appears that the original membranous structure of the rough surfaced endoplasmic reticulum has been retained as a part of the envelope of the organelle.

Unfortunately, we failed to induce experimentally the formation of these organelles in chicken fibroblasts and, although numerous cells of various tissue cultures have been investigated, these observations have been made only once. Histochemical tests thus could not be applied in order to determine the nature or function of these inclusions. We feel, however, that this brief report may aid further studies on this subject.

### SUMMARY

Morphological studies have been made on a functionally unknown organelle in cultured normal chicken fibroblasts and fibroblasts infected with Rous sarcoma virus. An organelle described by other investigators as being spindle-shaped has been contrasted with the rod-like fiber we have found which consists of a bundle of approximately 50 single fibrils, about 50 A in diameter, showing no cross-banding, and being loosely enclosed by a sheath or membrane. The width of the organelle is approximately 600 A; the length could not be determined. The fibers may extend throughout the entire cell, apparently in any direction. The organelle may occur singly or in bundles or in both forms, and seems not to be bound to any specific cell area. It is suggested that these structures are formed by the rough-surfaced endoplasmic reticulum. Their contents may be a phagocytized or secretion product of the cell. The formation of the organelles is not a result of infection by Rous sarcoma virus.

This investigation was supported in part by research grants CA-04692 of the National Cancer Institute and IN46D of the American Cancer Society, Inc. Received for publication, April 22, 1963.

### REFERENCES

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Abbreviations

n, nucleusm, mitochondriaf, fibers

fl, fibrils
fm, fiber membrane
er, endoplasmic reticulum

FIGURE 1 Portion of a chicken fibroblast grown in tissue culture, showing oblique and longitudinal sections through rod-like fibers. Arrow points to a portion of rough-surfaced endoplasmic reticulum as part of the envelope of the fiber. Lead stained. × 36,000.



Figure 2 Cross-section of a bundle-like pattern of fibers in fibroblasts. Lead stained.  $\times$  180,000.

Figure 3 Longitudinal section of single fibers in fibroblasts. Lead stained.  $\times$  180,000.

