Virus-Like Particles in Myeloma Cells of Man.* (29810)

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Although many animal tumors contain intracellular viruses which are etiologically related, observations of viral particles within human neoplastic cells have only been reported in rare and usually isolated cases(3,5). Recently, several investigators utilizing viral isolation techniques have indicated a more consistent association between viruses and some human tumors(4,6,7). In 1961 numerous intracytoplasmic particles, with the ultrastructural characteristics of viruses, were observed in neoplastic plasma cells from a patient with multiple myeloma(8). Recently, apparently identical intracellular particles have been observed in the myeloma cells from 2 similar patients with typical clinical features of this disease.

Case I. G.U., 66-year-old white man was first admitted to the hospital in Dec., 1963, with a history of back pain for 2 months. Roentgenograms demonstrated osteolytic lesions in his skull, ribs and vertebrae. The bone marrow contained numerous plasma cells and many of these possessed round cytoplasmic inclusions, variable in size up to 7 μ in diameter, which appeared pink to colorless in Wright stained material (Fig. 1). Serum globulins were elevated to a total of 3.8 g and the major portion was determined by electrophoresis to be Beta globulin. He was treated with local x-irradiation, chlorambucil and prednisone with only temporary symptomatic remission and he died with pneumonia 2 months after his initial hospital admission. Cytoplasmic inclusions were observed within myeloma cells in sections of bone marrow obtained at autopsy which were identical to those previously found in the bone marrow smear.

Case II. B.T., 57-year-old white woman was first admitted in Sept., 1963, with a history of anemia for several months. Roentgenograms were normal. Her bone marrow contained greatly increased numbers of plasma cells and a diagnosis of multiple myeloma was made. A few of the myeloma cells contained cytoplasmic inclusions identical to those in the previous patient. Serum globulins were elevated to 6.5 g and by electrophoresis they were mainly Beta in type. Immunoelectrophoresis revealed the abnormal globulin to be Beta 2A. She was treated with chlorambucil or phenylalanine mustard (double blind therapeutic protocol) with little objective effect. In March, 1964, the cytology of the bone marrow was essentially unchanged and the same type of cytoplasmic inclusion was present in some plasma cells at this time which was 6 months after the initial observation.

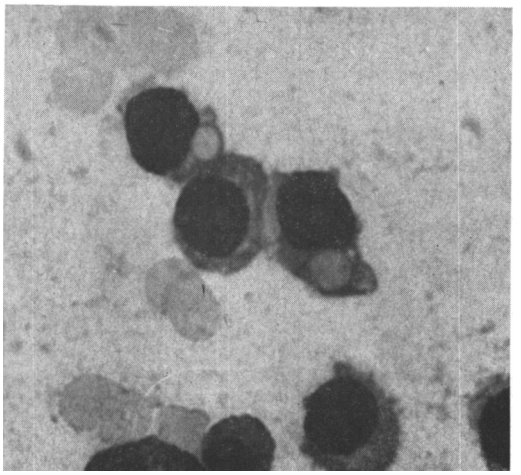

FIG. 1. Light micrograph of 2 myeloma cells with cytoplasmic inclusions × 900.

By electron microscopy cytoplasmic inclusions were readily found in myeloma cells from the bone marrow of Case I. Most of the inclusions appeared as large vacuoles surrounded by 2 closely spaced membranes (Fig. 2). Characteristically small round particles occurred about the edge of the vacuoles but not in the center. Less commonly smaller vacuoles filled with particles were observed. The rounded (occasionally ovoid) particles were uniformly from 50 to 70 mμ in diameter.

* This investigation was supported in part by USPHS Research Grants A1-04889-05 and GM 08574-02.

Most of the particles were doughnut forms with concentric double membranes and a central area with little electron density (Bernhard's type A)(1). A small number of the particles had a dense central core about 15-20 mμ in diameter with an appearance consistent with that of a nucleoid (Fig. 3). Such particles were more commonly found in the

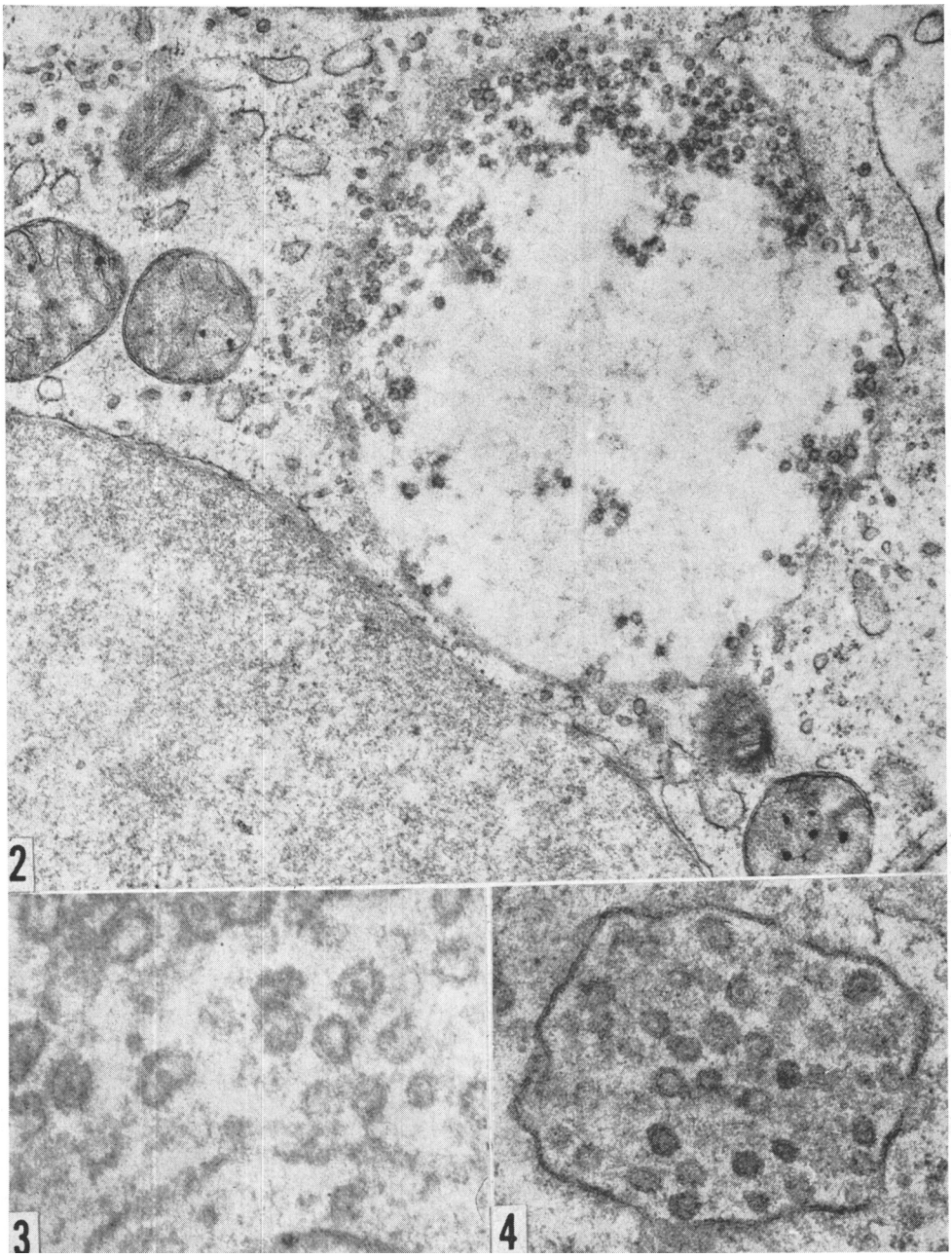

FIG. 2. Electron micrograph of myeloma cell with cytoplasmic vacuole containing small, rounded particles × 30,000.

FIG. 3. Particles, 50-70 mμ in diameter, within a portion of a vacuole similar to that in Fig. 2. Some of the particles possess a small, dense, central core. Case I. × 110,000.

FIG. 4. Particles within a cytoplasmic vacuole of a myeloma cell from Case II. × 70,000.

smaller aggregation of particles rather than at the edge of the larger vacuoles. At times fine radial projections were apparent at the periphery of the particles. Identical cytoplasmic vacuoles and particles have been observed by electron microscopy in myeloma cells from Case II (Fig. 4). These inclusions are present in the bone marrow specimen obtained in Sept., 1963, as well as in the material obtained in March, 1964.

The cytoplasmic vacuolar inclusions in these 2 patients with multiple myeloma appear identical to each other as well as to those described in the previous case(8). In all 3 cases the vacuoles contained apparently the same kind of particles with the ultrastructural characteristics of virus. It is also of interest that the serum protein abnormalities in these 2 cases as well as in the original patient are exactly alike, *i.e.,* a major increase in Beta globulin. Inclusions were prominent in many of the myeloma cells in Case I (G.U.) but in Case II (B.T.) and the original case the inclusions were uncommon by light microscopy and relatively difficult to find by electron microscopy. Although it is hard on only a morphologic basis to make a definite identification of these particles as viruses, they do possess general ultrastructural characteristics which are essentially the same as known viruses(2,5). Inclusions appearing identical to these were reported recently in 2 cases of lymphosarcoma(10). The authors suggested several possible origins for them including that they may be (1) a degenerative change, (2) related to secretion, (3) enlarged multivesicular bodies or (4) viral in development. All of these remain possible explanations for the inclusions described here.

Neoplastic plasma cells from 15 other patients with multiple myeloma have been observed by electron microscopy and similar inclusions have not been observed(10). Such negative results do not permit a determination of the incidence of such particles because of the sampling difficulties inherent in electron microscopy. The inclusions in the second case (B.T.) were not observed in the specimen obtained early in the course of her disease until a more extensive second search was made. Inclusions of this type have not been observed in the examination of bone marrow from 10 patients with sideroblastic anemia although plasma cells are frequently increased in this disease(9). However, the significance of this negative result in non-myeloma patients is also difficult to assess.

The similar appearance of the inclusions and particles in all 3 cases would strongly suggest some specific relationship between these particles which appear to be viruses and the myeloma cells. Although these may be related to subclinical viral infections in these patients, the identical particle type in all 3 patients makes this less likely. The appearance of the same type of particle in Case II (B.T.) at 2 intervals, 6 months apart, also would be against a coincident infection. However, the possibility that these particles represent a passenger virus in these myeloma cells has not been excluded. On the other hand the indicated specificity in their relationship to neoplastic plasma cells, *i.e.,* the same type of particle in the same type of tumor in 3 different patients with identical serum protein abnormalities would suggest that if these particles are viruses they may have an etiologic relationship to multiple myeloma.

Summary. Electron microscopy of bone marrow revealed cytoplasmic particles within neoplastic plasma cells from 2 additional patients with multiple myeloma and increased Beta globulin in their serum. Although a definite identification cannot be made, the ultrastructure of these particles in general is consistent with some known viruses.

1. Bernhard, W., Cancer Res., 1958, v18, 491.
2. ———, ibid., 1960, v20, 712.
3. ———, Prog. Exp. Tumor Res., Karger, Basel/New York, 1963, v3, 1.
4. Dalldorf, G., Bergamine, F., Proc. Nat. Acad. Sci., 1964, v51, 263.
5. Dmochowski, L., Prog. Exp. Tumor Res., Karger, Basel/New York, 1963, v3, 35.
6. Inman, D. R., Woods, D. A., Negroni, G., Brit. Med. J., 1964, v1, 929.
7. Negroni, G., ibid., 1964, v1, 927.
8. Sorenson, G. D., Exp. Cell Res., 1961, v25, 219.
9. ———, Am. J. Path., 1962, v40, 297.
10. ———, Lab. Invest., 1964, v13, 196.
11. Vasquez, C., Pavlovsky, A., Bernhard, W., Compt. Rend. Acad. Sci., 1963, v256, 2261.

Received September 2, 1964. P.S.E.B.M., 1965, v118.