BRIEF NOTE

Detection of Coronavirus-Like Particles in a Spontaneous Case of Feline Infectious Peritonitis

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(Received for publication July 5, 1977)

While feline infectious peritonitis (FIP) can be experimentally produced in cats by inoculation with cell-free materials from spontaneous cases [11], attempts at isolating the causative agent using conventional cell cultures have failed [4, 7]. In tissues of experimentally infected kittens, many particles resembling those of coronaviruses in morphology [1, 3, 6-11] and specific immunofluorescence were seen in cultured peritoneal cells derived from the experimentally infected kitten [7]. However, no reports have described the presence of the viral particles in tissues from naturally infected animals, while Osterhaus et al. [3, 6] demonstrated negatively stained virions in peritoneal exudate. This brief communication is to describe detection of coronavirus-like particles in the omentum and peritoneal macrophages of a clinically as well as pathologically diagnosed case of FIP.

A 7-month-old male mongrel cat prominently emaciated with slightly enlarged abdomen was moribund and subjected to euthanasia. No detailed clinical examination was performed, while signs of illness had been noted since 2 weeks. At autopsy about 500 ml yellowish-brown viscid ascites was present in the peritoneal cavity, being clotted when exposed to the air. The whole surfaces of the omentum, mesenterium and peritoneal organs were covered with considerable amount of greyish-white fibrinous deposit, forming focally multiple fine nodules. The omentum was severely thickened and edematous showing turbid appearance. Hepatic lobules were adhered with each other as well as with the peritoneum. Mesenteric lymph nodes showed edematous swelling, and the renal capsule was severely edematous. The pleura was also edematous with greyish-white multiple fine nodules with a slightly increased amount of yellowish-brown viscid exudate.

Sections were prepared from 10% formalin-fixed tissues of the serosa and main organs and they were stained with hematoxylin and eosin. A number of degenerative and necrotic foci with nuclear debris as well as metaplasia and hyperplasia with syncytium formation were seen in the...
mesothelium of not only the peritoneum, omentum, mesenterium and pleura but also the capsules of the liver, spleen, pancreas, digestive tract and mesenteric lymph nodes. There was focal or diffuse infiltration of macrophages, lymphocytes, plasma cells and occasionally neutrophils. The whole surface of the serous membranes was covered with a layer of fibrin deposit, and there were some parts which were replaced by either immature granulation tissues or fibrous connective tissues. Adventitial and perivascular infiltration of mononuclear cells was notable in smaller veins, arteries and lymphatic vessels. In the spleen and mesenteric lymph nodes, activation of reticuloendothelial cells with accumulation of plasma cells was seen. Edema and diffuse hyperplasia of reticuloendothelial system were present in the bone marrow. Pulmonary edema and catarrhal bronchopneumonia were found, but no significant changes were detected in the kidneys and central nervous system.

A portion of the omentum as well as pellet samples of ascites after low-speed centrifugation were fixed with 5% glutaraldehyde phosphate buffer, pH 7.2 at 4°C for 6 hr, rinsed with the same buffer, postfixed in 1% osmium tetroxide at 4°C for 2 hr and embedded in Epon 812. Ultrathin sections were made from these materials and stained with uranyl acetate and lead nitrate. Examination was made using electron microscopes, JEM 100S at 80 Kv and Hitachi HU-12 at 75 Kv.

A small number of virus particles, resembling those reported in experimental cases [1, 7–11], were found singly or in groups within cytoplasmic vesicles of peritoneal macrophages as well as mesothelial lining cells. Macrophages with the particles were enlarged, having degenerated mitochondria, proliferated microfilaments and lamellar bodies, well-developed phagosomes, distended endoplasmic reticulums and Golgi cisternae. The particles were seen within smooth-surfaced cisternae, the Golgi apparatus, lysosomes and phagosomes (Figs. 1 to 3). They were mostly spherical and 90 to 138 nm in diameter with club-shaped surface projections 15 to 20 nm in length (Fig. 2). The nucleoid 60 to 95 nm in diameter was doughnut-shaped with an electron lucent area at the center. The particles within the phagosomes and lysosomes were mostly degenerated. Occasionally, budding profiles were recognized from the smooth-surfaced endoplasmic reticulum but never from the plasma membrane (Fig. 2). No other types of particles identified as microorganisms were detected.

From a considerable amount of characteristic ascites, severe fibrinous inflammation and vasculitis in the serosa [2, 5], the present case is considered to be of a typical FIP. It is of interest that the viral particles were detected in large number within mesothelial cells, and the present observations have further confirmed the etiologic role of coronavirus in FIP, as already suggested from the results of experimentally infected cases [1, 7–11]. In the present case, the samples might have been collected during a stage favorable to detect viral particles. Nevertheless, the complete viral particles were not so large in number as described in experimental cases [8, 11], retaining a possibility that the virus might play only a triggering role for producing the disease entity. From the present case, transmission of the disease to kittens resulting in propagation of viral particles have been successful and the pathogenesis of FIP is being studied.
References


Explanation of Figures

Fig. 1. Coronavirus-like particles within a mesothelial cell of the omentum. Bar=200 nm.

Fig. 2. Coronavirus-like particles within the endoplasmic reticulum cisternae of a mesothelial cell. Bar=500 nm.

The insert shows particles having club-shaped projections in the cisternae of smooth surfaced endoplasmic reticulum (small arrow), one of which is probably in budding process (large arrow). Bar=100 nm.

Fig. 3. Coronavirus-like particles with a dense nucleoid within dilated cisternae of smooth-surfaced endoplasmic reticulum, Golgi apparatus, phagosomes and lysosomes of a macrophage. Bar=100 nm.