An Ultrastructural Observation of Esophageal Lesion in Bovine Papular Stomatitis

Hiroyuki M. OKADA, Yutaka CHIHAYA, and Kiyoshi MATSUKAWA
Department of Veterinary Pathology, School of Veterinary Medicine, Rakuno Gakuen University, Ebetsu, Hokkaido 069, Japan
(Received 31 October 1986/Accepted 9 May 1987)

KEY WORDS: bovine papular stomatitis, esophagus.

Bovine papular stomatitis (BPS) is a viral disease caused by a parapoxvirus and has been recognized in Japan. The lesions of BPS develop in the lips, oro-nasal mucosae, tongue and muzzle, and sometimes in the esophagus, rumen, reticulum and omasum [2, 3]. Clinical as well as pathological findings of natural cases of BPS have been published [2, 3, 8, 9, 11]; however, there have been no reports on ultrastructural study of the esophageal lesions.

A 10-year-old Holstein-Friesian dairy cow was clinically diagnosed as having traumatic pericarditis and died on May 21, 1984 [1]. At autopsy, multi-focal necrotic erosions and ulcers were seen on the mucosae of the tongue and esophagus (Fig. 1). The sizes of the lesions varied from rice grain to thumb-tip, their shape was ovoid and they were occasionally fused together. The margin of the lesions was whitish in color and raised. Tissues were fixed in 10% formalin and embedded in paraffin and glycol methacrylate (GMA)-Quetol 523M. Sections were cut and stained with hematoxylin and eosin (HE). Histopathologically, the mucosae around the necrotic erosions or ulcers were parakeratotic. Infiltration of lymphocytes and neutrophils was observed in the propria. The most characteristic change was hydropic degeneration of the epithelia noticed in the margin of lesions. All of the epithelial cells were swollen, and perinuclear edema was obvious. Nuclei were pyknotic and hyperchromatic in their margin, and occasionally contained an inclusion body stained deep purple in color. The cells also contained one or two basophilic or amphophilic cytoplasmic inclusions of various shapes and spherical eosinophilic inclusions (Fig. 2). The spherical eosinophilic inclusions measured from 1 to 20 μm in diameter, and occasionally attached to or were included in the basophilic or amphophilic inclusion. The fine granular eosinophilic networks were also observed around the basophilic or amphophilic inclusion.

For electron microscopy, formalin fixed specimens were postfixed in 1% osmic acid and embedded in Quetol 651. Ultrathin sections were stained with uranyl acetate and lead citrate. Serial sections about 1 μm thick were cut and stained with HE or toluidine blue for light microscopy. Many mature viral particles were scattered in the edematous cytoplasm. The basophilic or amphophilic inclusions of various shapes observed under the light microscope consisted of a granular matrix as well as immature and mature viral particles (Figs. 2 and 3). Immature viral particles were spherical in shape, 250 to 300 nm in diameter, and surrounded by a circular membrane (Fig. 4). Mature viral particles measured 340 by 170 nm in diameter and had the characteristics of parapoxvirus [10]. The spherical eosinophilic inclusions consisted of an

![Fig. 1. Multi-focal necrotic ulcers in the esophagus.](image-url)
electron-opaque granular matrix (Figs. 2–4). Ribosomal particles were often accumulated around the surface of electron-opaque inclusions (Fig. 4). The nuclei of infected cells occasionally contained tubular structures of 85 nm to 130 nm in the outer diameter and 35 nm to 72 nm in the inner diameter (Fig. 5). The present case was pathologically diagnosed as BPS.

The basophilic or amphophilic inclusions seen here may correspond to the B-type inclusions described by Kato et al. [5]. The granular matrixes containing mature and immature viral particles may represent viroplasm [6, 8, 9, 11]. On the other hand, the spherical eosinophilic cytoplasmic inclusions appeared similar to the A-type inclusions [5]. The A-type inclusions have been recognized in cow pox [4] as well as parapoxvirus infection, i.e. BPS [8], contagious pustular dermatitis [7], and seal pox [9]. Ribosomal clusters were observed at the surface of the A-type inclusions in cow pox [4]. Electron-opaque inclusions observed in the present case might correspond to the A-type inclusions. The fine eosinophilic granular networks in the cytoplasm may correspond to the scattered mature viral particles.

Intracytoplasmic tubular components seemingly corresponded to the intranuclear inclusions observed under the light microscope. Two types of intranuclear components were previously reported [7–9, 11]; one is tubular and the other filamentous. In the present case, only tubular components were observed. It is still unknown why two types of intranuclear components appeared in the nuclei of infected cells, though it was speculated that they might be a by-product of viral replication [11].

The present study clarified an association of ulcerative lesions of the esophagus with viral replication. Cytoplasmic inclusions have been histologically found in the epithelial cells of the fore stomach of ruminants infected with parapoxvirus [2, 3, 6]. Further ultrastructural studies may be required to clarify whether parapoxvirus replicates in the fore stomach of ruminants.
BOVINE PAPULAR STOMATITIS

Fig. 4. Viroplasm (V) contains immature viral particles as well as spherical electron-opaque granular matrixes (arrows). Ribosomal particles gather around the electron-opaque inclusion (arrowheads). Many mature viral particles in the cytoplasm. ×11,200.

Fig. 5. Tubular components (arrowheads) in the nucleus and mature and immature viral particles in the cytoplasm. ×14,000.

REFERENCES

5. Kato, S., Hagiwara, K., Baba, E., Sato, Y., and


要 約

ウシ奇疹性口炎罹患牛における食道病巣の超微形態的観察（短報）：岡田洋之・千早 豊・松川 清（酪農学園大学家畜病理学教室）——乳牛のウシ奇疹性口炎例の食道病巣を電顕的に観察した。空胞化した上皮細胞には2種類の細胞質亜核体、すなわち好塩基性あるいは両染性の不定形亜核体ならびに好酸性球状亜核体がみとめられた。前者は未成熟ならびに成熟ウイルス粒子を含む基質から成り、後者は高電子密度の顆粒状物であった。成熟ウイルス粒子は大きさ340×170nmで、パラボックスウイルスの特徴を示し、ウイルス粒子の認められた細胞の核内には、管状構造物がしばしば認められた。