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recovered. ASV was not recovered from, and antibodies were not detected in, the remaining 4 puppies. Of these, one had died after ICHV viremia, one was positive in ICHV detection, one was positive only in ICHV antibody, and another escaped the ICHV infection.

The above results may indicate that the ASV replicates *in vivo* only when ICHV has been co-infected. Helper dependent replication of ASV in dogs was thus clarified.

PATHOLOGICAL OBSERVATIONS OF THE BURSA OF FABRICIUS IN MAREK'S DISEASE

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In order to clarify the patho-morphological characteristics of the bursa of Fabricius (BF) in Marek's disease, chickens infected with Marek's disease virus (MDV-JM strain) were studied by light and electron microscopy. Ninety birds of 4 groups were examined: They consisted of 42 birds of a normal control group (4 to 157 days old), 20 birds of an inoculated group (on the 3rd to 54th day postinoculation), 16 birds of a contact exposure group (on the 3rd to 156th day postexposure) and 12 birds of a field group (103 to 168 days old) of Marek's disease (MD).

The BF of the normal control group grew with age and the maximum size was attained after 107 days old. Involuting changes (physiologic involution) were first observed on 157 days old after the beginning of egg-laying.

The BF involved 3 forms of lesions as follows:

- A; Disappearance of the follicles due to necrosis of the medulla (accompanied with inclusions in the inoculated group)
- B; Atrophy and disappearance of the follicles due to cyst-formation
- C; Neoplastic proliferation of the lympho-reticular cells in the interstitial tissue of the BF.

In the inoculated group, precipitating antibodies to MDV antigens were negative, and all the BF showed severe atrophy (pathologic involution). A-form lesions were found frequently and were severe, and B-form lesions were also observed. Viral antigens by immunofluorescent antibody techniques were localized frequently in the follicles. Intranuclear and cytoplasmic inclusions were found in the reticulum cells and epithelium-like cells in the medulla of the

follicles. By electron microscopy, herpes-type unenveloped virus particles were found in the nuclei and the cytoplasm of these cells, and enveloped particles were rarely present in the perinuclear and intercellular spaces.

In the contact exposure group, C-form lesions were observed and involuted changes were also found in the later stages.

In the field group of MD, C-form lesions were predominant and B-form lesions were also seen.

The histopathogenesis of the necrosis and disappearance of the follicles in the BF of chickens infected with MDV was discussed. By electron microscopy, the characteristics of the pathologic involution of the BF and morphological relationship between inclusions and herpes-type virus particles could be demonstrated.

A STUDY OF THE MULTIPLICATION OF THE AVIAN ENCEPHALOMYELITIS VIRUS IN CHICK PANCREATIC CELL CULTURES

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1) Cells obtained by collagenase treatment from chick pancreases were cultured, and then monolayers were made about 10 days after cultivation. The monolayers consisted of some sorts of epithelioid cells and fibroblasts at a low percentage.

2) The maximum virus titer of the cell-culture fluid ($10^{2.9}$ EID₅₀/ml) was obtained 8 days after having been inoculated with $10^{5.0}$ EID₅₀ of an embryo-adapted AEV (AEV-VR strain). The virus titers of the cell phase did not rise in parallel with those of the culture fluids. The infected cells maintained by medium changes gave virus titers of $10^{3.7}$ EID₅₀/ml in the cell-culture fluid 17 days after inoculation (AI). Virus multiplication was evidently observed in the inoculum of $10^{3.0}$ EID₅₀, but not in case of $10^{1.0}$ EID₅₀.

3) A chick-pancreas-passed AEV (UP strain, inoculum of pancreas suspension of $10^{3.6}$ EID₅₀) and a field isolate of AEV (K-71, inoculum of brain suspension of $10^{2.7}$ EID₅₀) multiplied in the cell cultures. The virus titers of the cell-culture fluids were $10^{3.0}$ and $10^{2.1}$ EID₅₀/ml respectively 8 days AI. The infected cell cultures maintained by medium changes for 15~20 days revealed virus titers of $10^{3.2}$ and $10^{2.9}$ EID₅₀/ml respectively. The UP strain multiplied more rapidly