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The detection of PCV 2 and PRRSV in the fattening pigs skin by immunohistochemistry
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Introduction and Objective

A newly emerged porcine disease syndrome, postweaning multisystemic wasting syndrome (PMWS), has been reported throughout North America and Europe and is associated with infection by a novel strain of porcine circovirus (PCV). Several experimental studies have suggested that coinfection with Porcine reproductive and respiratory syndrome virus (PRRSV) potentiates PCV-2 infection, resulting in PMWS. PMWS was diagnosed by the detection of characteristic circoviral inclusion bodies in affected tissues, viral antigen by immunohistochemistry, viral particles by electron microscopy, or isolation of the virus. The purpose of this study was to describe the detection of PCV-2 and PRRSV in the fattening pigs skin by immunohistochemistry.

Materials and Methods

Piglets were considered to have PMWS with the following specific lesions: weight loss or wasting, histopathological lesions of PCV-2 infection, and detection of PCV-2 antigen within lesions in affected tissues by IHC testing.

We used formalin-fixed, paraffin-embedded sections of mesenteric lymph node and skin to localize PCV-2 antigen with the use of a rabbit polyclonal antibody as previously described with slight modifications: 3- μ m thick sections were dried at 60°C for 10 minutes, deparaffinized and rehydrated routinely, quenched with 3% H₂O₂ for 5 minutes, rinsed in 2 changes of DW for 5 minutes, treated with 0.05% protease (Invitrogen, Germany) for 10 minutes, rinsed in 2 changes of DW for 5 minutes and in 0.01M TBS for 5 minutes at room temperature, and then treated with primary antibody (National Veterinary Research and Quarantine Service, Korea) at 1:1000 dilution for 30 minutes at room temperature. Slides were rinsed in TBS, treated with biotinylated secondary antibody (DAKO Corporation, USA) for 30 minutes, rinsed in TBS, treated with DAB (DAKO) for 5 minutes,

rinsed in 2 changes of DW, counterstained in instant hematoxylin (DAKO) for 2 minutes, washed DW for 5 minutes, dehydrated, cleared and coverslipped.

Results

Approximately 80% of the normal architecture is obscured and replaced by multifocal to coalescing aggregates of numerous macrophages with abundant eosinophilic cytoplasm (epithelioid) and fewer multinucleate giant cells with up to 20 nuclei that are either located along the periphery of the cytoplasm (Langhans type) or within the center (foreign body type). Immunohistochemically, PCV-2 antigen was demonstrated in piglets; antigen localized diffusely in the cytoplasm and to various sized, often multiple, and usually intracytoplasmic inclusions.

Reference

- W. G. Van Alstine, et al. Effect of formalin fixation on the immunohistochemical detection of PRRS virus antigen in experimentally and naturally infected pigs. *J. Vet Diagn Invest* 14:504-507(2002).
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