NOTE  Pathology

Congenital Cutaneous Fibropapillomatosis with No Evidence of Papillomavirus Infection in a Piglet

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ABSTRACT. Multiple yellowish-white, cauliflower-like mass lesions on the skin of the head and back in a 4-month-old piglet were pathologically examined. These lesions had developed before the weaning period. Histologically, the cutaneous neoplasms were characterized by papillary outgrowth of connective tissue covered by thick epidermis. Hyperplasia of the epidermis was corresponded with proliferation of capillaries, lympho-plasmacytic infiltration, and proliferation of fibroblasts in the dermal stroma. There were no inclusion bodies and significant degeneration in the keratinocytes. Papillomavirus antigen and DNA were not detected in these lesions by immunohistochemistry and polymerase chain reaction, respectively. Accordingly, the fibropapillomatosis of the present case might be hamartomatous rather than infectious.

KEY WORDS: dermatopathology, fibropapilloma, hamartoma, pathology, swine.


Cutaneous papilloma is rare in pigs, and two patterns of the lesion have been reported: transmissible genital papilloma and congenital fibropapilloma [7, 13]. Only two cases of porcine congenital papilloma have been reported, and the involvement of viral infection in the cutaneous lesion has not been confirmed [11, 16].

In animals and humans, some forms of cutaneous papilloma are caused by papillomavirus (PV). In particular, bovine papilloma virus (BPV) types 1, 2, and 5 induce fibropapilloma of the skin in cattle [4, 6]. Congenital papillomas are considered non-infectious lesions that most often occur in horses [5, 14, 17]. In a previous report on porcine congenital papilloma, the presence of virus was not found on electron microscopy, but there have been no studies using immunohistochemistry or polymerase chain reaction (PCR) to assess for viral involvement [16]. We recently encountered cutaneous papillomatosis in a piglet. In this report, we describe the histopathological features and the results of immunohistochemistry and PCR for PV.

A hybrid piglet developed multiple, yellowish-white, cauliflower-like tumors on the skin of the head and back. According to the owner, these lesions had been observed from the suckling period (Fig. 1), and the animal had not shown any other significant clinical abnormalities. The pig was sacrificed at 4 months of age and the liver, intestine, lymph nodes, and 4 tumors were submitted to our laboratory for pathological examinations. The largest tumor among those submitted was approximately 8.0 × 7.0 × 5.0 cm in size and was hard, creamy white, and pedunculated on cut section (Fig. 2).

Tissue samples were fixed in 10% neutral-buffered formalin and embedded in paraffin wax. The sections (4 μm) were cut and stained with hematoxylin and eosin (HE). For immunohistochemical examination, a standard peroxidase-antiperoxidase technique was used on the paraffin sections with primary antibodies as rabbit polyclonal anti-bovine papillomavirus (BPV-1) (DakoCytomation, Carpinteria, CA, U.S.A.) and mouse monoclonal anti-proliferating cell nuclear antigen (PCNA) (clone PC10; DAKO, Glostrup, Denmark). Before incubation with primary antibodies, deparaffinized sections were pretreated with 0.1% trypsin for 1 hr at 37°C and microwave heating in citrate buffer (pH

Fig. 1. Multiple cutaneous papillomas on the head (A) and back (B) at suckling period.
6.0) for 10 min at 90°C for PV and PCNA, respectively. Peroxidase-conjugated anti-mouse IgG [Histofine Simple Stain MAX-PO (M); Nichirei, Tokyo, Japan] and anti-rabbit IgG [Histofine Simple Stain MAX-PO (R); Nichirei] were used as secondary antibodies. The reaction products were visualized by 3,3′-diaminobenzidine tetrahydrochloride (Wako Pure Chemical Industries, Ltd., Tokyo, Japan) and the sections were counter-stained with hematoxylin. As a positive control for immunostaining of PV antigen, we used paraffin sections of canine cutaneous viral papilloma and canine pigmented viral plaque.

DNA was extracted from the paraffin sections as follows. Deparaffinized sections were suspended in a buffer containing 20 mg/ml proteinase K (Wako), 3.75 mol/l NaCl, 100 mmol/l Tris-HCl (pH 8.0), 500 mmol/l EDTA (pH 8.0), and sodium dodecyl sulfate, and were incubated at 37°C for 24 hr. DNA was then, extracted in phenol and chloroform-isoamyl alcohol (CIA, 24:1). Finally, about 1 μg of DNA sample was subjected to PCR using consensus primers MY11/09 (MY11: GCM CAG GGW CAT AAY AAT GG, MY09: CGT CCM ARR GGA WAC TGA TC). The PCR primer pair MY09/MY11 was originally designed to human PV types and has been reported to amplify the L1 gene of some animal PV types [2, 9]. DNA extracted from the paraffin sections of canine papilloma with canine PV-4 infection was used as a positive control and that of the intact porcine skin as a negative control.

Histologically, the cutaneous neoplasm was characterized by prominent papillary outgrowth of fibro-vascular dermal tissue and overlying thick epidermis (Fig. 3). The epidermis showed moderate laminated orthokeratotic hyperkeratosis. Colonized cocci were observed on the epidermal surface. Hyperplastic and rete peg-like extensions of the epidermis were observed in some areas and corresponded with the growth of capillaries, infiltration of lymphocytes and plasma cells, and proliferation of fibroblasts in the dermis. The keratinocytes did not contain giant keratohyalin granules, koilocytosis, blue-grey cytoplasm, intranuclear inclusion bodies, or mitotic figures (Fig. 4). By immunohistochemistry, PV antigen was clearly detected in the nuclei of affected keratinocytes in the tissues of canine cutaneous viral papilloma and canine pigmented viral plaque. However, there were no papillomavirus antigen-positive cells in the present case. Nuclear PCNA was detected predominantly in the basal layer of the overlying epidermis and occasionally in the fibroblasts and vascular endothelial cells in the superficial stromal tissue. In the consensus primer PCR, amplicons (450 bp fragments of L1 ORF) were not found in tissue samples (Fig. 5).

In previous reports of porcine congenital cutaneous papilloma, vertical infection of PV was suspected but not confirmed [11, 16]. In humans and cows, papilloma virus is known to cause vertical infection, and PV infection of the fetus has been confirmed by PCR [1, 12, 15]. However, the present case was PV-negative on immunohistochemistry and PCR.
In porcine PV infection, transmissible genital papilloma is rarely observed. This virus induces papilloma in the preputial diverticulum of pigs, after which the lesion begins to regress and eventually peel off [7]. Histologically, the lesion is characterized by extensive irregular acanthosis and epithelial outgrowth with no significant mesenchymal reaction. Moreover, the infected keratinocytes are swollen with acidophilic intracytoplasmic inclusions [13]. These PV-induced changes of keratinocytes are also found in dogs, cows, humans, and rabbits, and are considered to be the typical histological finding of PV infection [7]. In the contrast, the present case did not show any histological characteristic changes of PV infection such as giant keratohyalin granules, koilocytosis, blue-grey cytoplasm, or intranuclear inclusion bodies. PCNA was predominantly expressed in the basal layer of the overlying epidermis, as in the normal epidermis. Furthermore, there were no other cutaneous papilloma or fibropapilloma cases in the pig farm where the present case had been fed. Hence, viral infection is unlikely to remain as the cause of this fibropapilloma.

Noninfectious congenital papilloma occurs in humans and may be present on any part of the body, but most commonly on the side of the face [10]. This is in agreement with previous reports of a horse [8], pig [16], and cow [3]. In human cases, the lesion may be caused by the anomalous growth of tissues surrounding the first branchial cleft. Therefore, the ear is also morphologically abnormal in these cases. However, there are no reports of congenital papilloma among animals with abnormal ears. In addition, in an infant, multiple papillomas have been reported to result from adhesion of the skin to the amnion [10]. The results of PCR and immunostaining for PV in this study may indicate a hamartomatous-like etiology as the cause of porcine congenital fibropapillomatosis. To confirm the hypothesis, further studies using a large number of cases should be examined.

REFERENCES