Lymphoid Neoplasms in Swine

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ABSTRACT. Seventeen cases of lymphoid neoplasms in swine were investigated and divided into eight histological types. Cases 1–3 were precursor B lymphoblastic leukemias, which occurred in three piglets from the same dam. Cases 4 and 5 were diagnosed, respectively, as a precursor B lymphoblastic lymphoma and a thymic B cell lymphoma, because there were cytological differences between the lymphomas. These five cases of immature B cell malignancies expressed CD79a and terminal deoxynucleotidyl transferase (TdT). Mature B cell lymphomas were divisible into follicular (case 6), diffuse centroblastic (case 7) and intestinal large B cell (cases 8–11) lymphomas. Unlike in case 7, the neoplastic cells in cases 8–11 showed cytological features intermediate between centroblasts and immunoblasts. The mature lymphomas were characterized by positive immunolabeling for CD79a and cytoplasmic immunoglobulins. A case of thymic γδ T cell lymphoma (case 12) were positive for CD3, CD5, WC1 and TdT. Instead of TdT, perforin was expressed in γδ T cell lymphomas (cases 13–17), whose histological characteristics were epitheliotropism, homing into T cell zones of lymphatic tissues, and cytological atypia and pleomorphism. In the present study, lymphoid neoplasms could be classified into discrete histological types, some of which were considered to be specific for swine.

KEY WORDS: classification, lymphoblastic leukemia, lymphoma, swine.


Lymphoma is the most frequently reported cancer of swine based on abattoir surveys [7]. Data on swine familial or hereditary lymphoma are available in few reports; in a herd, the occurrence of 53 cases of lymphoma in young pigs was associated with inbreeding [4], and in another report, two sows (dam and daughter) were affected with lymphoid neoplasms without splenomegaly and with prominent splenomegaly, respectively [20]. In contrast to these reports with no immunological information, a great number of thymic lymphomas found in the young female offspring of one bull were characterized by the expression of some B cell markers and terminal deoxynucleotidyl transferase (TdT), a marker for immature lymphocytes [2].

One or few histological types are presented in most immunohistochemical studies on swine lymphoid neoplasms [23, 24]. In one study, however, six cases were divisible into four histological types according to the World Health Organization (WHO) classification for humans [5]. In addition, signet ring cell lymphoma [13], lymphoepithelioid lymphoma [8] and T cell-rich B cell lymphoma [25] have been observed in swine, although they are relatively rare even in humans. This paper describes 17 cases of swine lymphoid neoplasms that were classified into eight histological types. Among them, three cases of leukemia of immature B cell origin, which occurred in siblings, were considered to be hereditary.

MATERIALS AND METHODS

Animals: Clinical and macroscopic findings in cases 1–7 and 12 are summarized in Table 1. All cases were mixed breed pigs. Three piglets (cases 1–3) were born in a pig farm. After mating with a boar, a 2-year-old sow (mother: Landrace; father: Large Yorkshire) delivered nine piglets on her second parturition, but one died at the age of 115 days (case 1). Although another Duroc boar was used for mating, two of 12 piglets obtained at the next parturition died at the age of 70 days (case 2) and 96 days (case 3). There was no genetic relationship between the boars. The cases were characterized by a rapid clinical course without specific signs. Except in the auricles of case 3, extranodal tumor formation was absent. Cases 4–17 were pigs brought to an abattoir in good condition. The animals were 6 months old in cases 8–11 and 13–17, and were females (cases 8 and 13–15) or castrated males (cases 9–11, 16 and 17). In cases 8–11, the small intestinal wall was highly thickened, and the intestinal or mesenteric lymph nodes were enlarged. In cases 13–17, enlargement of the bronchial, hepatic, renal, mesenteric or superficial lymph nodes was detected, and there were neoplastic lesions in the spleen, liver, kidneys or lungs.

Histology and immunohistochemistry: Tissue samples were fixed in 10% buffered formalin and embedded in paraffin. Tissue sections were stained with hematoxylin and eosin (HE) and Giemsa. Immunohistochemistry was applied to paraffin sections. Labeling was performed using
the streptavidin-biotin-peroxidase complex (SBC) method with SBC kits (Nichirei, Tokyo, Japan). The primary antibodies employed were: rabbit polyclonal antibodies to immunoglobulin M (IgM) (μ chain specific) and IgA (α chain specific) (Bethyl Laboratories, Montgomery, TX, U.S.A.), to κ light chain and λ light chain (BioGenex Laboratories, San Ramon, CA, U.S.A.), and to CD3 (Dako A/S, Glostrup, Denmark), CD5, a marker for T lymphocytes (Lab Vision, Fremont, CA, U.S.A.) and TdT (Dako Corporation, Carpinteria, CA, U.S.A.); goat polyclonal antibody to IgG (Fc specific) (Bethyl); and mouse monoclonal antibodies to CD79a (Dako A/S), WC1, a marker for γδ T lymphocytes (AbD Serotec, Oxford, U.K.) and perforin (Sumitomo Electric Industries, Osaka, Japan).

RESULTS

Histological findings: In all cases, neoplastic growths were detected in the macroscopically visible lesions. All cases, except case 6, displayed a diffuse growth pattern. Histological and cytological characteristics were closely similar in cases 1–3, and the architecture of the lymph nodes examined and spleen was completely obliterated by leukemia cells. There were numerous intravascular leukemia cells in the lungs and kidneys. In case 1, the thymus was invaded by neoplastic cells, but the thymic architecture was preserved. The leukemia cells were medium-sized to large, with small to moderate amounts of cytoplasm. The nuclei were round, oval or slightly irregular, with mild to moderate clumping of chromatin, and there were occasional large nucleoli.

In case 4, there were massive neoplastic growths in the skeletal muscles and kidney. Intravascular neoplastic cells were few or absent in the organs examined. Except in localized neoplastic lesions, no neoplastic cells were detected in the bone marrow. The neoplastic cells resembled those in cases 1–3, but larger cells predominated (Fig. 1A). In case 5, the normal architecture of the thymus was completely replaced by sheets of lymphoma cells. The cells were small to large in size, and had round to oval nuclei with fairly condensed chromatin and inconspicuous nucleoli, and there was little cytoplasm (Fig. 1B).

In case 6, follicle center-like structures were observed throughout the tissues of some lymph nodes, but diffuse neoplastic growths were detected in others. Neoplastic invasion into lymph follicles was detected in cases 7 and 9. The neoplastic cells in case 6 were moderate or large in size, with round to oval nuclei, slightly to moderately condensed chromatin and small to moderately prominent nucleoli. There was relatively little cytoplasm (Fig. 1C). In case 7, the neoplastic cells resembled the large cells in case 6 (Fig. 1D). In cases 8–11, the neoplastic cells showed morphology intermediate between that of centroblasts and immunoblasts. The cells resembled those of case 7 in nuclear shape and nucleolar size and location, but the chromatin was less condensed. The cytoplasm tended to be more abundant than that of case 7 but not significantly so (Fig. 1E).

In case 12, the neoplastic cells resembled those in the thy-

<table>
<thead>
<tr>
<th>Case</th>
<th>Sex</th>
<th>Age</th>
<th>Clinical findings</th>
<th>Gross pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>C</td>
<td>115 days</td>
<td>Underdeveloped and emaciated. The animal became depressed and somewhat anorexic, and the next day showed dysstasia and died</td>
<td>Enlargement of the kidneys, with densely distributed petechiae on the surface. Slight enlargement and hemorrhage of the parotid, mandibular and superficial inguinal lymph nodes. Multiple subcutaneous petechiae</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>70 days</td>
<td>Sudden depression and death</td>
<td>Slight enlargement and edema of the spleen, liver and lungs. Densely distributed subcutaneous petechiae</td>
</tr>
<tr>
<td>3</td>
<td>F</td>
<td>96 days</td>
<td>Sudden death</td>
<td>Enlargement of the spleen, and superficial cervical, superficial inguinal, bronchial and internal iliac lymph nodes. Multiple grayish white foci in the auricles of the heart. Mild enlargement of the liver. The carcass appeared slightly pale</td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>6 months</td>
<td>No abnormal findings</td>
<td>Tumor masses in skeletal muscles and in the kidneys</td>
</tr>
<tr>
<td>5</td>
<td>C</td>
<td>6 months</td>
<td>No abnormal findings</td>
<td>A mediastinal tumor mass. Enlargement of the mediastinal, hepatic and internal iliac lymph nodes</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>&gt;2 years</td>
<td>No abnormal findings</td>
<td>Enlargement of abdominal lymph nodes. Neoplastic lesions in the spleen and kidneys</td>
</tr>
<tr>
<td>7</td>
<td>F</td>
<td>6 months</td>
<td>No abnormal findings</td>
<td>Enlargement of the hepatic and mesenteric lymph nodes. Small nodules on the surface of the spleen and liver</td>
</tr>
<tr>
<td>12</td>
<td>F</td>
<td>6 months</td>
<td>No abnormal findings</td>
<td>Enlargement of the thymus and spleen</td>
</tr>
</tbody>
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C = castrated male; F = female.

Table 1. Clinical data and gross pathology in cases 1–7 and 12
Fig. 1. Histology of neoplastic cells. (A) Case 4, kidney. Large neoplastic cells predominate, and there are some large nucleoli. HE. Bar=5 μm. (B) Case 5, mediastinal tumor. The lymphoma cells are characterized by inconspicuous nucleoli and small amounts of cytoplasm. HE. Bar=5 μm. (C) Case 6, lymph node. The neoplastic follicle center is composed of centrocytoid and centroblastoid cells, and the former tend to have more condensed chromatin. HE. Bar=5 μm. (D) Case 7, lymph node. The neoplastic cells show similar cytology to that of the centroblastoid cells in Fig. 1C. HE. Bar=5 μm. (E) Case 11, small intestine. Most neoplastic cells display more widespread cytoplasm than do centroblastoid cells. HE. Bar=5 μm. (F) Case 12, mediastinal tumor. As in Fig. 1B, the neoplastic cells have inconspicuous nucleoli and relatively scant cytoplasm. HE. Bar=5 μm. (G) Case 16, splenic lymph node. The neoplastic cells show considerable pleomorphism and atypia. HE. Bar=5 μm.
mic B cell lymphoma (case 5) (Fig. 1F). In case 13, the thymus was heavily infiltrated by neoplastic cells. Tropism of neoplastic cells was observed in bronchial and duodenal (case 13), pyloric (case 14), interlobular bile duct (cases 16, 17), and renal pelvic (case 15) epithelia, and homing into T cell zones was in the spleen (cases 13–17) and lymph nodes (cases 14, 15). There were reactive lymphatic follicles in the neoplastic tissues of the lungs and thymus (case 13), pylorus (case 14), liver (case 16), kidneys (case 15) and lymph nodes (cases 14, 15, 17). The neoplastic cells were highly variable in size. The nuclei were vesicular with irregular contours, and prominent nucleoli were frequently seen. The cytoplasm was moderate to abundant in amount (Fig. 1G).

**Immunohistochemical findings:** The results and diagnoses are shown in Table 2. CD79a (Fig. 2A) and TdT (Fig. 2B) were expressed in cases 1–5, and CD79a and cytoplasmic IgM (cIgM) (Fig. 2C) or cytoplasmic IgG (cIgG) (Fig. 2D) were detected in cases 6–11. Neoplastic cells in case 12 and in cases 13–17 were positive respectively for CD3, CD5, WC1 (Fig. 2F) and perforin (Fig. 2G).

**DISCUSSION**

In the WHO classification, neoplasms of immature B cell origin are precursor B lymphoblastic leukemia or lymphoma [9]. In the current study, five cases of immature B cell neoplasms expressing TdT were detected. Because lumina of the pulmonary blood vessels were filled with neoplastic cells, cases 1–3 were diagnosed as leukemia [15, 16]. In contrast, cases 4 and 5 were characterized by formation of tumor masses, and were diagnosed as lymphoma. As in bovine precursor B cell neoplasms [15, 17], there were cytological differences between precursor B lymphoblastic leukemia/lymphoma (cases 1–4) and thymic B cell lymphoma (case 5), and the latter was regarded as an independent histological entity.

Da Costa et al. [2] found a total of 216 cases of thymic B cell lymphoma in the young female offspring of one bull, and the cases were characterized by TdT expression. Similarly, a great number of lymphomas appeared in the offspring of a number of different sows that had been mated to the same boar and, in addition, there was a close kinship between the animals [4]. Such a widespread occurrence is due to the imbalance between the size of the populations of boars and sows. In contrast, sow-related genetic disorders such as in cases 1–3 occur on a smaller scale, and are more difficult to detect. The preponderance of very young animals with familial precursor B cell neoplasms suggests that genetic factors are an important cause of precursor lymphoid neoplasms.

A diagnosis of follicular lymphoma was made in case 6, because there were follicle center-like structures throughout the neoplastic tissues of some lymph nodes. This finding implies that the neoplastic cells are capable of forming such structures [9]. In contrast, replacement of lymphatic follicles by lymphoma cells was observed in case 7, and a diagnosis of diffuse centroblastoid lymphoma was made [11]. Like in the previously reported cases of follicle center cell origin [11], small amounts of IgM production were detected in the cases.

Although IgA is the major isotype produced in the gastrointestinal tract, IgM has an important role in mucosal immunity in swine [3] and IgG-producing cells predominate in inflammatory lesions of the intestine [18]. In cases 8–11, the lymphoma tissues located chiefly in the small intestine were composed of cells intermediate between centroblasts and immunoblasts [11, 12], and the neoplastic cells were frequently positive for cIgG. Similar cytological features have been reported in ileal lymphomas in young pigs, though not only cIgG-positive but also cIgM-positive cases were detected [23]. These cytological and immunohistochemical findings suggest that this type of lymphoma is a distinct disease entity [23], and a diagnosis of intestinal large B cell lymphoma was made. The preferential occurrence of the lymphoma in young pigs may be due to the facts that piglets are prone to diarrhea [27] and have an immature immune system.

The neoplastic cells in case 12 were similar to those in case 5, and were less pleomorphic than those in peripheral T cell lymphomas, including cases 13–17 [5]. In a normal thymus, WC1-positive lymphocytes are present chiefly in the medulla, whereas TdT-positive ones are observed exclusively in the cortex (unpublished data). Both markers were detected in case 12, and the lymphoma cells were considered to be at the transitional stage from cortical thymocytes to medullary thymocytes. Because γδ T lymphocytes are thymic dependent in swine and abundant in the thymic medulla [19], WC1 expression in thymic lymphomas is presumably not unusual. In a previous report of thymic T cell

| Case | CD79a | λ | κ | μ | γ | α | CD3 | CD5 | WC1 | perforin | TdT | Diagnosis                  |
|------|-------|---|---|---|---|---|-----|-----|-----|-----|-----|----|---------------------------|
| 1–3  | +     | - | - | - | - | - | -   | -   | -   | -   | -   | + | Precursor B lymphoblastic leukemia |
| 4    | +     | - | - | - | - | - | -   | -   | -   | -   | -   | + | Precursor B lymphoblastic lymphoma  |
| 5    | +     | + | - | - | - | - | -   | -   | -   | -   | -   | + | Thymic B cell lymphoma           |
| 6    | +     | + | - | - | - | - | -   | -   | -   | -   | -   | + | Follicular lymphoma             |
| 7    | +     | + | - | - | - | - | -   | -   | -   | -   | -   | - | Diffuse centroblastoid lymphoma  |
| 8–11 | +     | + | + | + | + | + | -   | -   | -   | -   | -   | + | Intestinal large B cell lymphoma |
| 12   | -     | - | - | - | - | - | +   | +   | +   | +   | + | Thymic γδ T cell lymphoma        |
| 13–17| -     | - | - | - | - | - | +   | +   | +   | +   | + | γδ T cell lymphoma              |

++: mostly or frequently positive, +: occasionally or rarely positive; −: negative.
Fig. 2. Immunohistochemistry of neoplastic cells. (A) Case 1, mesenteric lymph node. CD79a is expressed in the cytoplasm of leukemia cells. SBC. Bar=5 μm. (B) Case 5, mediastinal tumor. Many lymphoma cells exhibit TdT nuclear staining of varying intensity. SBC. Bar=5 μm. (C) Case 6, lymph node. Some neoplastic follicle center cells show cytoplasmic μ heavy chain expression. SBC. Bar=5 μm. (D) Case 7, lymph node. Large neoplastic cells are positive for γ heavy chain, but the nuclei are devoid of centrally located large nucleoli. SBC. Bar=5 μm. (E) Case 12, mediastinal tumor. Tumor cells of varied size frequently show surface staining for CD5. SBC. Bar=5 μm. (F) Case 14, lymph node. Several WC1-positive tumor cells are visible. One of them possesses an irregularly contoured nucleus (arrow), and is readily distinguishable from normal lymphocytes. SBC. Bar=5 μm. (G) Case 13, pancreaticoduodenal lymph node. Most neoplastic cells have dot-like perforin-positive deposits in the cytoplasm. SBC. Bar=5 μm.
lymphoma, the neoplastic tissue was composed mainly of medium-sized cells [10], which expressed TdT but not WC1 (unpublished data), and this case is interpretable as showing the thymic cortex phenotype.

Bovine γδ T cell lymphoma is characterized by epitheliotropism in different types of epithelia, homing into T cell zones, and expression of WC1 and perforin [14, 22]. Since similar features were observed, cases 13–17 were diagnosed as γδ T cell lymphoma. Considering the high proportion of γδ T lymphocytes in swine peripheral blood [21], this type of lymphoma, as in cattle [6], is probably not rare. Despite such similarities between the bovine and swine cases, the neoplastic cells were far more pleomorphic in the swine cases. Likewise, the neoplastic cells are pleomorphic in human nonhepatosplenic γδ T cell lymphomas that involve either skin, intestine or nasal regions, but vary in size among patients [1, 26].

The WHO classification of human lymphoid neoplasms is not directly applicable to bovine neoplasms and requires certain modifications [6, 9, 17]. The current study disclosed the presence of four distinct entities—thymic B cell lymphoma, intestinal large B cell lymphoma, thymic γδ T cell lymphoma and γδ T cell lymphoma with tropism for various epithelia—that are not listed in the human classification. Taking into account that there are species differences in epithelia—that are not listed in the human classification.


