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SHORT COMMUNICATION Clinical Case Report

Pericardial mesothelioma with severe congestive heart failure in a Holstein cow

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Abstract

A 58-month-old Holstein cow showed anorexia, edema of the lower jaw and dewlap, jugular venous engorgement, abdominal gas, and watery diarrhea. Heart sounds were faint on auscultation, and decreased rumen motility was noted in physical examination. Echocardiography findings included adhesion of fibrin-like structures to the pericardium, highly echogenic periaortic region, and pericardial effusion, which suggested traumatic pericarditis or tumor formation. Although atypical mesothelial cells were observed in bloody pericardial fluid, no diagnosis was made. At complete necropsy, milky mass formation was observed on the epicardium. Histopathological examination led to a diagnosis of primary malignant pericardial mesothelioma.

Key Words: congestive heart failure, Holstein, pericardial mesothelioma

Mesothelioma is a neoplasia originating from the pleural, pericardial membrane, or peritoneal mesothelial cells, and observed commonly in calves[14,15,24]. Several reports have described peritoneal mesotheliomas in cattle, but less information is available on the clinical aspects of mesotheliomas of pericardial origin[4,19,22]. The present case report describes ante-mortem findings, including clinical signs, ultrasound and cytological features, and hematological and biochemical examinations of malignant mesothelioma originating from the pericardial membrane in an adult cow with severe congestive heart failure. Clinical diagnosis of mesothelioma in cattle is also discussed.

A 58-month-old Holstein dairy cow was brought to a local veterinarian with chief complaints of anorexia and decreased milk production 6 months after normal delivery. On Day 1, physical examination revealed severe edema of the lower jaw and dewlap, jugular venous engorgement, increased abdominal gas, and watery diarrhea. Neostigmine was administered for decreased rumen motility as symptomatic treatment. However, the general condition of the cow worsened, with faint heart sounds on auscultation on Day 2.

On Day 4, the cow was transferred to the Animal Teaching Hospital at the Obihiro University of Agriculture and Veterinary Medicine. On initial

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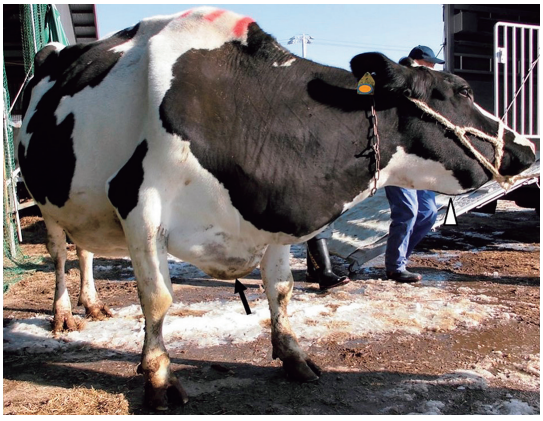

Fig. 1. Edema of the lower jaw (white arrow head) and dewlap (black arrow) were apparent on Day 4.

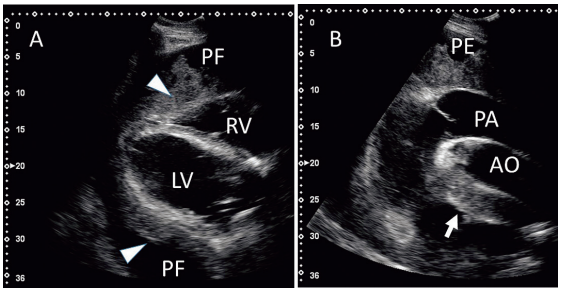

Fig. 2. Echocardiography (A) at ventricular level; pericardial fluid and adhesion of fibrin-like structures (white arrow heads) to the pericardium were observed. PF: pericardial fluid, RV: right ventricle, LV: left ventricle. (B) At aortic level; pleural effusion and highly echogenic periaortic region (arrow) were observed. PE: pleural effusion, PA: pulmonary artery, AO: aorta.

physical examination at the hospital, high rectal temperature (40.4°C), tachycardia (120 beats/min), and polypnea (36 breaths/min) were noted. Edema of the lower jaw and dewlap, jugular venous engorgement, and watery diarrhea noted on Day 1 were also observed (Fig. 1). Heart sounds were very faint on auscultation, and percussion of the thoracic wall suggested fluid retention resulting in pleural or pericardial effusion. The lower voltage of each wave in the electrocardiogram also suggested pleural/pericardial effusion.

Echocardiography revealed pericardial fluid and adhesion of fibrin-like structures to the pericardium (Fig. 2A). Pleural effusion and highly echogenic periaortic region were also observed (Fig. 2B). Blood-like liquid was recovered from both pericardial and thoracic cavities by thoracentesis. Analysis of the pericardial fluid revealed a red blood cell count (RBC) of 1.75×10^6/μl, white blood cell count (WBC) of 5,500/μl, total protein (TP) of 2.2 g/dl, and specific gravity (SG) of 1.022. A sediment smear of this blood-like pericardial fluid showed clusters of round to polygonal cells with pleomorphic nuclei and aggregated nucleoli. The cells varied in size and appeared to weakly adhere to each other (Fig. 3). Reddish pleural effusion with an RBC of 0.62×10^6/μl, WBC of 1,400/μl, TP of 0.8 g/dl, and SG of 1.015 was obtained from the right thoracic cavity. Pleural effusion with an RBC of 1.47×10^6/μl, WBC of 2,700/μl, TP of 1.8 g/dl, and SG of 1.020

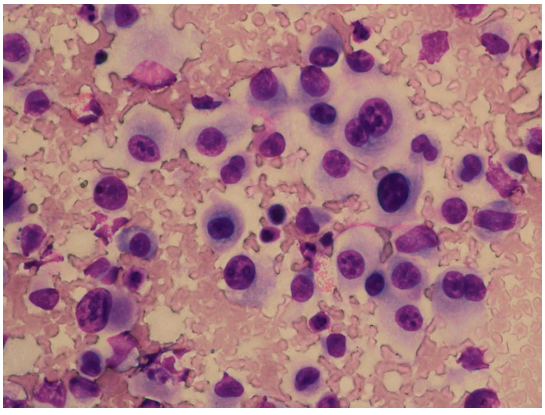

Fig. 3. Sediment smear of blood-like pericardial fluid showed clusters of round to polygonal cells with pleomorphic nuclei and aggregated nucleoli. The cells varied in size and appeared to weakly adhere to each other (x400, Giemsa stain).

was also recovered from the left thoracic cavity. Similar cells as observed in the pericardial fluid smear were observed in the smear of the pleural effusion. Clear and yellowish ascites was collected and found to be transudate with a TP of 0.9 g/dl, SG of 1.015, and WBC of 300/μl.

The results of hematological examinations are summarized in Table 1. A mild neutrophilia with normal WBC, and mildly increased gamma-glutamic transferase activity were noted, as well as slight decreases in total protein and albumin concentrations and A/G ratio. Both LDH and thymidine kinase activities were within normal range. Bovine leukemia virus infection was

Table 1. Results of hematological and biochemical examinations (Day 4)

Test	Result		Normal range	Reference	Test	Result		Normal range	Reference
RBC	5.68	$\times 10^6$/μl	5.0–7.2	3)	BUN	12.5	mg/dl	10–25	12)
Hb	9.6	g/dl	8.6–11.9	3)	Creatinine	0.62	mg/dl	0.4–1.0	12)
HCT	27.5	%	23.1–31.7	3)	ALP	130	U/l	23–78	12)
WBC	9,700	/μl	5,600–12,700	3)	γ-GTP	90	U/l	< 40	12)
Sta	388	/μl	50–720	3)	LDH	1,078	U/l	697–1,445	12)
Seg	5,626	/μl	1,100–5,700	3)	Thymidine kinase	2.5	U/l	< 5.4	20)
Lym	1,261	/μl	2,300–9,300	3)	Total protein	6	g/dl	7.2–9.0	12)
Mon	776	/μl	0–600	3)	Albumin	2.5	g/dl	3.2–4.0	12)
Eos	1,649	/μl	0–2,000	3)	A/G ratio	0.58	g/dl	0.86–1.18	12)
Platelets	3,120	$\times 10^3$/μl	210–710	3)					

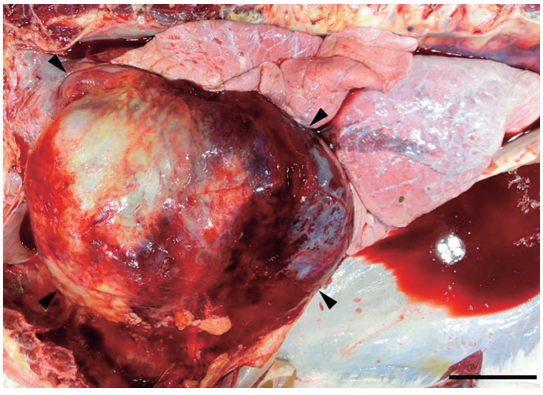

Fig. 4. Gross region of the thoracic cavity. The pericardium is enlarged (arrow head) and occupies the thoracic cavity. Bar = 10 cm.

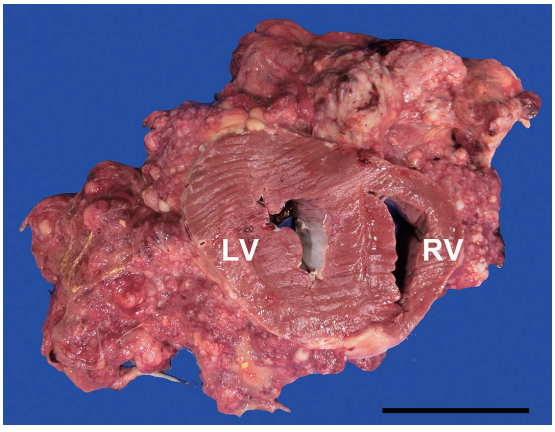

Fig. 5. Lateral section of the heart with masses. The epicardium is covered with irregular masses. RV: right ventricle. LV: left ventricle. Bar = 10 cm.

evaluated with a commercial enzyme-linked immunosorbent assay kit (Enzootic Bovine Leukosis ELISA Kit, JNC, Tokyo, Japan) and found to be negative.

Although approximately 10 l of pericardial fluid, a total of 12 l of pleural effusion from both thoracic cavities, and 15 l of ascites were removed by aspiration on Day 5, the general condition worsened over the next few days, and the cow was euthanized on Day 8.

At necropsy, the thoracic cavity contained a large amount of hemorrhagic pleural effusion and enlarged pericardium (H 40 cm × W 30 cm × D 30 cm) (Fig. 4). The pericardial cavity contained blood-like effusion and fibrins. The pericardium and epicardium were severely thickened and covered with a number of white-to-yellowish masses. The masses were 1–5 cm in diameter, confluent and elastic, and it was difficult to remove them from the epicardium (Fig. 5). At the cut surface, hemorrhage and necrosis were observed in some parts of the masses. Other gross findings included increments of yellowish clear peritoneal effusion, severe edema in subcutaneous tissues and mesentery, and chronic congestion of the liver. Tissue samples were fixed in 15% neutral-buffered formalin and embedded in paraffin. Paraffin sections were stained with Hematoxylin and Eosin (HE). Immunohistochemistry was performed using monoclonal anti-human cytokeratin (clone AE1/AE3, Dako, Denmark) and monoclonal anti-vimentin (clone V9, Dako, Denmark) antibodies. The simple stain MAX-PO polymer reagent

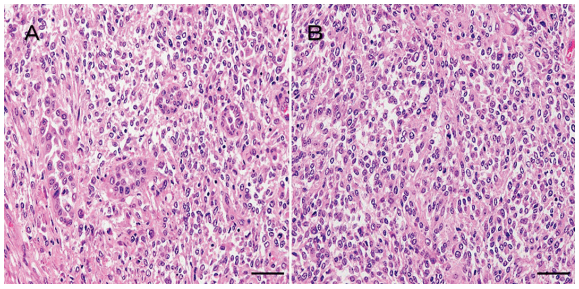

Fig. 6. Histological features of neoplastic cells. (A) Epithelial-like cells arranged in a tubular pattern. (B) Mesenchymal-like cells arranged in a lace-like pattern. Hematoxylin and eosin stain. Bar = 200 μm.

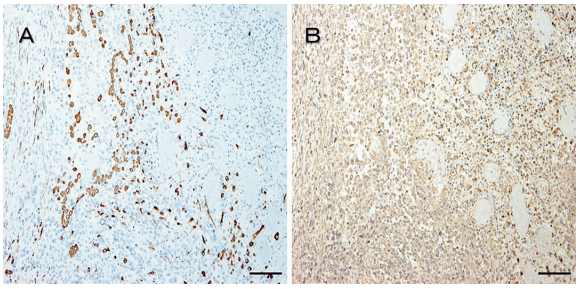

Fig. 7. Immunohistochemistry of neoplastic cells. (A) Epithelial-like cells and some mesenchymal-like cells were immunopositive for cytokeratin. (B) Neoplastic cells were diffusely immunopositive for vimentin. Hematoxylin counterstain. Bar = 100 μm.

(Nichirei Bio-science, Tokyo, Japan) was used as the secondary antibody.

Histological examinations revealed the proliferation of neoplastic cells and infiltration of inflammatory cells. Neoplastic cells were pleomorphic, exhibited a round, polygonal, or short spindle shape, and had eosinophilic cytoplasm and oval nuclei with polymorphism. Some neoplastic cells were epithelial-like, lining the epicardium or arranged in a tubular or papillary pattern. Other neoplastic cells were mesenchymal-like, proliferating as connective tissue and arranged in a lace-like pattern (Fig. 6). Mitotic figures were readily encountered. Moreover, metastatic regions were observed only in accessory lymph nodes but not in any other organs. Immunohistochemically, the epithelial-like cells were immunopositive for cytokeratin and vimentin. Most of the mesenchymal-like cells were immunopositive for vimentin, and some were also immunopositive for cytokeratin (Fig. 7).

Necropsy and histopathological examinations led to a definitive diagnosis of primary malignant pericardial mesothelioma. It was speculated that increased bloody pericardial effusion produced by pericardial mesothelioma caused diastolic heart dysfunction, followed by congestive heart failure symptoms including edema, pleural effusion, and ascites.

In the initial diagnosis, traumatic pericarditis was suspected based on the clinical findings of fever and severe congestive heart failure and echocardiographic images with the deposition of fibrin-like structures on the pericardium and pericardial effusion[11,16]. The additional finding of bloody pericardial fluid and the results of hematological and biochemical examinations showing non-severe inflammation eliminated the possibility of traumatic pericarditis. Although these findings were suggestive of possible idiopathic pericardial fluid, this was ruled out based on the echocardiography and cytology findings[5,9,16]. Neoplastic diseases such as lymphoma, hemangiosarcoma, and mesothelioma were also suspected given the highly echogenic periaortic area observed in echocardiography[16,25]. As atypical mesothelial cells were found in the pericardial and pleural effusion, mesothelioma was thought to be the most likely diagnosis in the present case.

A differential diagnosis between a reactive and neoplastic proliferation of mesothelial cells by cytology is generally difficult to make in small animals and humans, as a wide variety of morphological changes are also observed in reactive proliferation of mesothelial cells, such as irregularity in nucleus size and atypical nuclei[1,7,8,13–15,17]. In the present case, clusters of atypical mesothelial cells were observed in cytology of pericardial fluid, but it was impossible to distinguish between reactive and neoplastic mesothelial cells. A recent study in dogs showed that only 7.7% were diagnostic by cytologic analysis of pericardial effusion[1]. Immunohistochemical analysis has been shown to be useful in

differentiating reactive mesothelial cells from malignant mesothelioma in humans[13], and thus, these methods can be applied for the diagnosis of mesothelioma in veterinary medicine.

Both LDH and TK activities are known as serum biomarkers of bovine leukosis[10,20,23]. In the present study, lower activities of both LDH and TK were useful for ruling out bovine leukosis. Cytokeratin fragments and mesothelin have recently been reported to serve as useful serum biomarkers for malignant mesothelioma in humans[6,21]. The usefulness of these biomarkers for accurately diagnosing bovine mesothelioma should be evaluated in the future.

In conclusion, clinical aspects including ultrasound and cytological features, and laboratory findings are useful for the ante-mortem diagnosis of malignant mesothelioma in cattle.

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