Successful Diagnosis of Type II Enteropathy-associated T-cell Lymphoma Using Flow Cytometry and the Cell Block Technique of Celomic Fluid Manifesting as Massive Pyoid Ascites that Could Not Be Diagnosed via Emergency Laparotomy

Hiroaki Tanaka1, Satoshi Ambiru1, Shunta Nakamura1, Terumi Itabashi2, Seiji Furuya2, Takenori Shimura2, Yuhei Nagao1, Chika Kawajiri2, Yusuke Takeda3, Shinichiro Hashimoto1 and Chiaki Nakaseko3

Abstract

Enteropathy-associated T-cell lymphoma (EATL), an intestinal tumor of intraepithelial T lymphocytes, is a rare and highly aggressive disease. We herein describe a case of type II EATL with massive pyoid ascites in which a histological examination could not be performed despite emergency laparotomy that was successfully diagnosed using flow cytometry and the cell block technique to analyze the celomic fluid. This case suggests that EATL should be included in the differential diagnosis of pyoid ascites of unknown origin and that flow cytometry and the cell block technique of assessing celomic fluid are useful procedures for diagnosing EATL, especially in cases in which conducting a histological examination is impossible.

Key words: enteropathy-associated T-cell lymphoma, flow cytometry, cell block technique, ascites, emergency laparotomy


Introduction

Serous effusion is a common complication of malignant lymphoma that frequently provides an important clue for diagnosis, especially in cases in which conducting a histological examination is impossible. Unlike pleural effusion, ascites is a rare presentation of malignant lymphoma (1).

Enteropathy-associated T-cell lymphoma (EATL), an intestinal tumor of intraepithelial T lymphocytes, is a rare and highly aggressive disease with a poor prognosis (2-7). EATL can be subdivided into two groups according to morphological findings, immunophenotypes and the association with celiac disease (4-6). Type I EATL, the classical form of EATL, is the predominant form and is associated with celiac disease, which is highly frequent in Northern Europe. Type II EATL, which was newly classified in the latest World Health Organization criteria for the classification of lymphoid neoplasms, occurs sporadically and is not associated with celiac disease (2). Type II EATL is very rare, and only a few reports of this disease are available (8, 9). The clinical course of EATL is highly aggressive. Since intestinal obstruction and perforation are common, many patients require emergency laparotomy and are diagnosed based on histological examinations of the resected tumors (3, 6). If a tumor specimen from emergency laparotomy is not available for a histological investigation, it is difficult to diagnose EATL because most EATL cases are limited to the gastrointestinal tract and mesenteric lymph nodes, while other lymph nodes and lymphoid tissues, such as the superficial
lymph nodes and bone marrow, are usually not involved (4, 5).

We herein describe a case of type II EATL with massive pyoid ascites that could not be diagnosed via emergency laparotomy, but rather was successfully diagnosed using flow cytometry and the cell block technique to analyze the celomic fluid.

**Case Report**

A 66-year-old woman who had received left nephrectomy for renal carcinoma was admitted to our hospital due to massive ascites. She presented with abdominal distension and anorexia that had started three weeks previously. She had no history of diarrhea or malabsorption and did not complain of abdominal pain, diarrhea or constipation at the time of admission. A physical examination performed on admission revealed that the patient’s abdomen was severely distended but soft with no tenderness, and her bowel sounds were weak. Her legs showed severe bilateral edema. There was no superficial lymph node swelling or skin lesions. The laboratory findings were as follows: white blood cell count, 14.5x10^9/L (without abnormal cells); hemoglobin, 11.7 g/dL; platelet count, 279.0x10^9/L, total protein, 4.4 g/dL; albumin, 1.3 g/dL; lactate dehydrogenase, 349 IU/L; C-reactive protein (CRP), 28.47 mg/dL; soluble interleukin 2 receptor, 3,930 IU/mL; CA125, 141.6 U/mL; and TPA, 172 U/mL. Serological tests for human immunodeficiency virus, human T-cell lymphotropic virus type I, hepatitis B virus and hepatitis C virus were all negative. A bone marrow examination revealed no abnormal cells. Culture tests of the ascites fluid detected Klebsiella pneumoniae and Candida glabrata. Moreover, obtaining a cytodiagnosis of the ascites fluid was not possible, as the cells had disintegrated due to bacterial contamination. Abdominal and pelvic computed tomography (CT) performed at the time of admission showed massive ascites and intestinal tract expansion. However, there was no free air, increased intestinal wall thickness, intestinal tumors, lymphadenopathy or cirrhosis of the liver (Fig. 1).

The patient was tentatively diagnosed with idiopathic peritonitis, and antibiotics and intravenous nutrition were started. However, seven days after admission, emergency laparotomy was performed because an abdominal CT scan obtained following the exacerbation of abdominal distension showed free air. At the time of laparotomy, there was a massive amount of pyoid ascites. The omentum had dissolved and rigidly adhered to the transverse colon and small intestine. The entire intestinal tract was severely edematous and agglomerated, with tissues adhering to each other. The small intestine in the left abdomen was perforated by a 1-cm hole that could not be resected for histological examination due to severe adhesion and was instead only sutured. We were unable to determine the cause of the intestinal perforation (Fig. 2a, b). Lavage and drainage were performed, and intravenous hyperalimentation was started. An upper and lower gastrointestinal series performed after the laparotomy revealed that there were no areas of perforation in the esophagus, stomach or colon. Eight days after the laparotomy, as a result of the antibiotic therapy and drainage, the pyoid ascites fluid in the drainage tubes gradually became serous and the CRP level decreased to 3.32 mg/dL. However, the amount of ascites fluid in the drainage tube remained over 1,200 mL/day, and bilateral pleural effusion developed and rapidly increased. We performed a cytodiagnosis and flow cytometry of the ascites fluid, which became serous after the laparotomy and development of the pleural effusion. The fluids contained atypical lymphoid cells (Fig. 3a) that were positive for CD3 (97.8%), CD7 (98.7%), CD8 (95.0%), CD56 (96.9%) and CD103 (93.9%) and negative for CD4, CD5, CD19 and CD20 (Fig. 3b). The remaining materials in these fluids were made into cell blocks for the histomorphological and immunohistochemical studies, which revealed medium-sized abnormal monomorphic cells with denuding or fine granular and hyperchromatic nuclei (Fig. 4a) that were CD3+ CD56+ CD5+ CD20+ on immunochemical staining (Fig. 4b-e). The cells were negative for Epstein-Barr virus-encoded small RNA on in site hybridization (EBERISH) (Fig. 4f). The patient was diagnosed with type II disease after taking her clinical course into consideration.

She suffered from respiratory failure caused by the pleural effusion, and mechanical ventilation was required. The hypoalbuminemia deteriorated as a result of protein loss in the massive ascites and pleural effusion. In addition, disseminated intravascular coagulation (DIC) was observed. THFCOP chemotherapy (pirarubicin 30 mg/m², day 1; vin-
cristine 1.4 mg/m², day 1; cyclophosphamide 750 mg/m², day 1; prednisolone 80 mg/body, days 1-5) was started. The patient’s general status improved temporarily; however, during the cytopenia phase, she suffered from melena, sepsis and deterioration of DIC and died of multiple organ failure. We were unable to perform an autopsy.

**Discussion**

In patients with type II EATL, the lymphoma cells are monomorphic, ranging from small to medium in size, in contrast to the pleomorphic medium to large cells observed in patients with type I EATL. An inflammatory background is usually present in patients with type I EATL but absent in those with type II EATL (2, 6). Immunophenotypically, the lymphoma cells in type II EATL patients are CD8⁺CD56⁻ (4-6, 9), distinct from type I EATL cells, which typically have a CD3⁺CD4⁻CD8⁻CD56⁻/⁺ phenotype (4-6).

In both type I and type II EATL patients, CD103 is a very useful marker for making the diagnosis of EATL, as it has been shown to be expressed on intraepithelial lymphocytes (10). It is notable that, in our case, there were CD103-

---

Figure 2.  a: At the time of laparotomy, the omentum had dissolved and rigidly adhered to the transverse colon and small intestine. The whole of the intestinal tract was severely edematous and agglomerated, with tissues adhering to each other. b: The small intestine in the left abdomen was perforated by a small hole. We were unable to identify the cause of the intestinal perforation.

Figure 3.  a: Atypical lymphocytes developed in the pleural effusion after the operation. b: A flow cytometry analysis of the pleural effusion fluid.
positive T-cell lymphoma cells in the pleural effusion and ascites fluid. Some case reports describe the usefulness of cytodiagnosis and flow cytometry of ascites fluid for diagnosing type II EATL (11, 12). The celomic fluid is informative because, in type II EATL patients, the lymphoma cells are monomorphic and an inflammatory background is absent (2, 6). Therefore, flow cytometry may be more useful for diagnosing type II EATL than type I EATL. Cell blocks prepared from residual tissue fluid can be used as adjuncts to smears in order to establish a more definitive cytopathological diagnosis (12, 13). In our case, the ascites fluid could not be evaluated on admission due to bacterial contamination; however, the ascites fluid in the drainage tube after laparotomy and the pleural effusion, which developed as a result of EATL progression, was very useful for making the diagnosis of EATL. According to EBER-ISH of the cell block specimen, our case was differentiated from NK/T-cell lymphoma, nasal type. Therefore, in cases in which conducting a histological examination is impossible, we recommend confirming the status of the celomic fluid using flow cytometry and analyses of cell blocks prepared from the residual tissue fluid to differentiate type II EATL from type I EATL or another type of lymphoma.

In patients with EATL, the most common presenting feature is abdominal pain, which is observed in over 80% of patients with EATL (3, 5). To our knowledge, there are few case reports of EATL with massive ascites. The massive ascites without abdominal pain observed in our case may be a rare presentation of EATL. It is possible that the hole that formed the intestinal perforation was very small and the pyoid ascites gradually effused without abdominal pain or free air. In addition, there may have been massive transudate effusion due to severe hypoalbuminemia before the perforation and peritonitis developed. EATL is an aggressive malignancy that invariably leads to death due to sepsis or peritonitis if left untreated (6). There may be other patients with massive pyoid ascites who have died of peritonitis or sepsis without receiving a diagnosis of EATL, despite undergoing emergency laparotomy.

In conclusion, we experienced a case of type II EATL that manifested as massive pyoid ascites without abdominal pain and was successfully diagnosed by analyzing the patient’s celomic fluid. Our case suggests that it is necessary to include EATL in the differential diagnosis of pyoid ascites of unknown origin and that, in cases in which conducting a histological examination is impossible, the use of flow cytometry and the cell block technique to analyze the celomic fluid is very helpful for making the diagnosis of EATL.

The authors state that they have no Conflict of Interest (COI).

Acknowledgement
The author would like to thank Takeichiro Kuwahara of the Chiba Foundation for Health Promotion and Disease Prevention for conducting the pathological analysis using the cell block
technique.

References


© 2014 The Japanese Society of Internal Medicine
http://www.naika.or.jp/imonline/index.html