J Infect Chemother 22 (2016) 815–818

Contents lists available at ScienceDirect

Journal of Infection and Chemotherapy

journal homepage: http://www.elsevier.com/locate/jic

Case Report

Paragonimus westermani infection mimicking recurrent lung cancer: A case report

Naoya Itoh [a, *], Mika Tsukahara [a], Hiroshi Yamasaki [b], Yasuyuki Morishima [b], Hiromu Sugiyama [b], Hanako Kurai [a]

[a] Division of Infectious Diseases, Shizuoka Cancer Center Hospital, Sunto-gun, Shizuoka, Japan
[b] Department of Parasitology, National Institute of Infectious Diseases, Shinjuku-ku, Tokyo, Japan

ARTICLE INFO

Article history:
Received 24 May 2016
Received in revised form
4 July 2016
Accepted 5 July 2016
Available online 5 August 2016

Keywords:
Paragonimiasis
Paragonimus westermani
Parasitic disease
Recurrent lung cancer

ABSTRACT

Herein, we report a case of *Paragonimus westermani* infection, which required differentiation from recurrent lung cancer. A 66-year old Japanese man with a history of lung cancer who had undergone a lobectomy was referred to our clinic for treatment of cough, sputum, dyspnea, and a right pulmonary nodule. He had previously eaten seafood he visited China. *P. westermani* infection was confirmed by the presence of antibody against *P. westermani* antigen in the patient's serum and eggs in his sputum. Eventually, molecular identification by PCR-restriction fragment length polymorphism analysis and sequencing confirmed that the patient was infected with triploid forms of *P. westermani*.

1. Introduction

Human paragonimiasis is a foodborne parasitic disease caused by infection with lung flukes of the *Paragonimus* species. In Japan, *Paragonimus westermani* and *Paragonimus miyazakii* are common pathogens, and human paragonimiasis is generally caused by *P. westermani* [1]. Moreover, in today's Japan, human paragonimiasis tends to be more prevalent among middle-aged men [2,3]. The symptoms and radiological findings of *P. westermani* are non-specific and varied. Therefore, human paragonimiasis may be misdiagnosed as pulmonary tuberculosis or lung cancer. We herein describe a case of *P. westermani* infection at a tertiary care cancer center during postoperative follow-up period for lung cancer that required differentiation from recurrent lung cancer.

2. Case report

A 66-year-old Japanese man with a history of squamous cell carcinoma of the right lower lung lobe, and who had undergone a right lower lobectomy 19 months before (Figs. 1A, B, 2A, B), was referred to our clinic because of progressive cough, sputum, dyspnea on exertion, and a nodular lesion on the right middle lobe. The symptoms had begun approximately 10 months earlier, and at the same time, left-sided pleural effusion was noted. Thoracentesis revealed yellowish, exudative pleural effusion with an increased level of eosinophils and no malignant cells. A nodular lesion (17 × 20 mm) on the right middle lung lobe had been detected 7 months earlier. The lesion continued to grow gradually in the subsequent 6 months. Although bronchoscopy was performed 1 month earlier, there was no evidence of malignancy. As there was no clinical improvement, he was referred to our clinic by his thoracic surgeon so that recurrent lung tumor could be ruled out and accurate diagnosis could be established. The patient had a medical history of hypertension and diabetes mellitus. His prescription medication included 10 mg/day nifedipine and 2.5 mg/day enalapril.

In a medical interview that included a detailed travel history, the patient reported that he had visited China on 3 occasions, 17, 13, and 8 months prior, and Vietnam on 2 occasions, 4 and 2 months before his visit to our clinic. On further questioning, he denied having experienced fever, rigors, night sweats, headaches, nausea, vomiting, chest pains, rashes, abdominal pain, and diarrhea. On physical examination, his blood pressure was 131/89 mmHg, heart rate was 82 beats/min, respiratory rate was 16 breaths/min, oxygen

* Corresponding author. Shizuoka Cancer Center Hospital, 1007 Shimonagakubo, Nagaizumi-cho, Sunto-gun, Shizuoka Prefecture, 411-8777, Japan. Fax: +81 55 989 5783.
E-mail address: itohnaoya0925@ybb.ne.jp (N. Itoh).

http://dx.doi.org/10.1016/j.jiac.2016.07.002

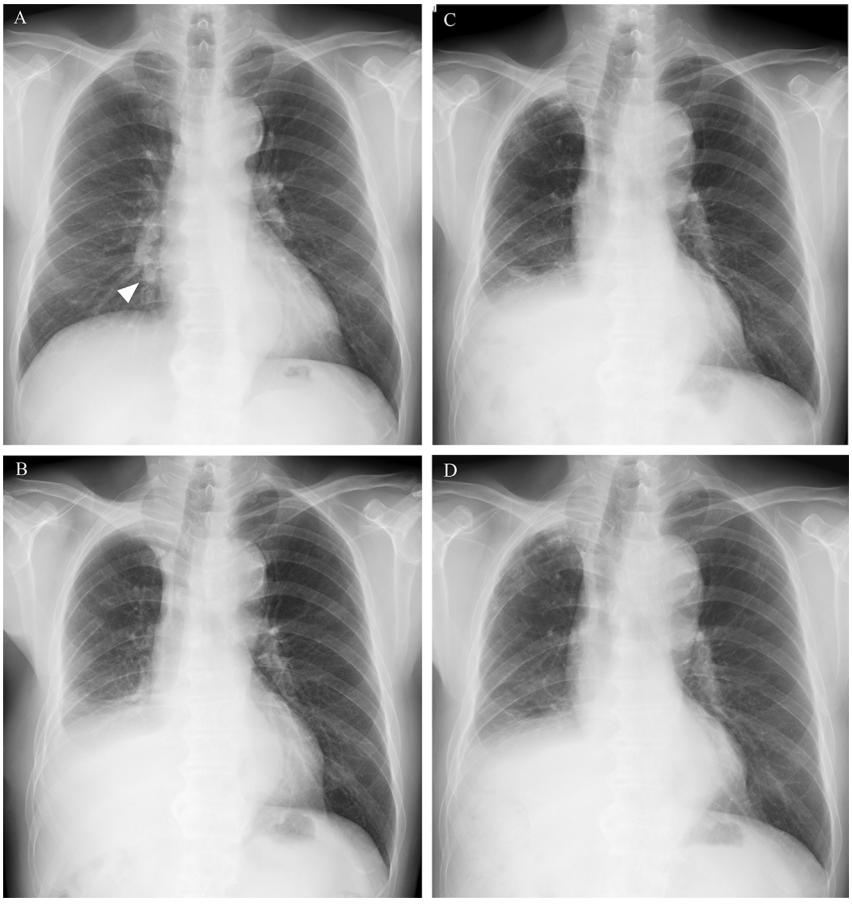

Fig. 1. Chest radiographs. (A) A chest radiograph 20 months prior shows a nodule (arrowhead) in the right lower lung field. (B) A chest radiograph taken after a right lower lobectomy 18 months before the consultation day shows no abnormality in the residual lung on the operative side. Compared with Panel B, chest radiographs on the consultation day (C) and 2 months after the completion of treatment with praziquantel (D) show no change.

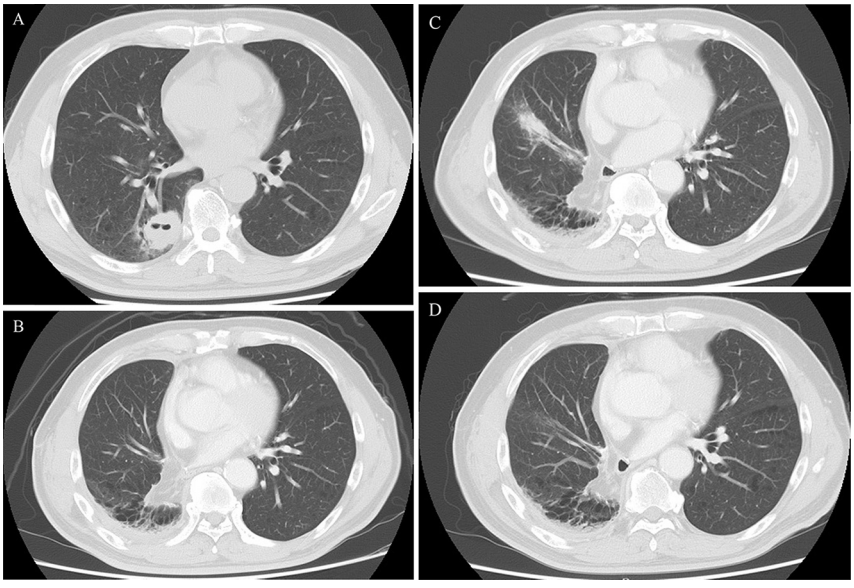

Fig. 2. CT of the thorax. (A) A CT scan 20 months before the consultation visit shows a mass lesion (36 × 34 mm) in the right lower lung lobe. (B) A CT scan after the right lower lobectomy, which was performed 16 months before the consultation visit, shows no abnormality. (C) A CT scan taken 3 weeks before the consultation visit shows a nodular lesion (24 × 22 mm) in the right middle lung lobe. (D) A CT scan at 5 months after the completion of treatment with praziquantel reveals no nodular lesion in the right middle lung lobe.

saturation while breathing ambient room air was 98%, and body temperature was 36.6 °C, respectively. Auscultation revealed decreasing respiratory sounds at the right anterior and right posterior lung zones. Superficial lymphadenopathy and hepatosplenomegaly were not detected. On laboratory examination (Table 1), the white blood cell count was 6980/μL with eosinophilia (13.6%). Eosinophilia was also seen 10 months earlier. The C-reactive protein level was 0.28 mg/dL. Serum tumor marker assessment showed normal carcinoembryonic antigen levels. While there was no abnormality on chest radiograph, computed tomography (CT) showed a nodular lesion (24 × 22 mm) in the right middle lung lobe (Figs. 1C and 2C) and no pleural effusion.

Although malignant cells were not identified in the sputum, trematode eggs, which were likely those of lung flukes, were found (Fig. 3). The eggs were oval with thickened shells at the non-operculated end. The patient recalled eating seafood on his visit to China but denied obvious ingestion of raw or improperly cooked freshwater crab, crayfish, or other foods known to harbor the parasite, such as wild boar meat and deer meat. The patient's serum was positive against *P. westermani* antigen and weakly positive against *Strongyloides ratti*, *P. miyazakii*, and *Fasciola hepatica* antigens by multi-dot enzyme-linked immunosorbent assays (ELISA). Finally, the patient was clinically diagnosed with *P. westermani* infection based on his symptoms, ova in his sputum, and anti-*P. westermani* antibodies. After the initiation of praziquantel (75 mg/kg for 3 days) treatment, his symptoms were promptly reduced accompanied by improvements in the nodular lesion (Fig. 2D) and peripheral blood eosinophilia. Eventually, molecular identification by PCR-restriction fragment length polymorphism (RFLP) analysis and sequencing confirmed that the ova were triploid forms of *P. westermani* (Fig. 4).

Table 1
Laboratory data.

Date	WBC/μL	Eosinophils (%)	CRP (mg/dL)	CEA (ng/mL)
12/26/2013[a]	9580	5.4	0.26	5.8
03/06/2014	7900	3,0	1.23	2.1
08/14/2014	7560	3.7	0.16	2.3
11/20/2014	8380	17.3	0.42	2.6
04/09/2015	8970	27.0	0.18	2.5
08/20/2015	7890	15.0	N/A	3.8
09/14/2015[b]	6980	13.6	0.28	N/A
10/27/2015	5500	6.5	0.14	N/A
11/25/2015	5640	2.0	0.15	N/A
02/04/2016	8310	3.1	N/A	N/A

WBC = White blood cell count; CRP = C-reactive protein; CEA = carcinoembryonic antigen.
[a] Before the day of operation.
[b] Consultation day.

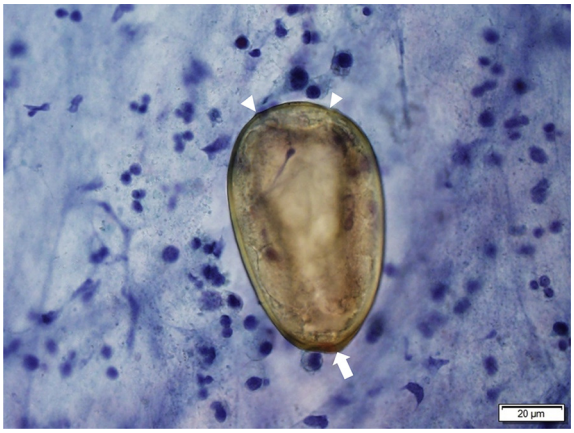

Fig. 3. Photomicrograph of an egg detected in a Papanicolaou-stained sputum specimen from the patient. The egg was ovoid in shape and golden brown in color, had an operculum at one end (between arrowheads), and measured 85 × 52 μm. The eggshell was thin on the lateral side, but the shell thickened at the non-operculated end (arrow). Based on the morphological characteristics of the egg, we tentatively identified the species as a triploid form of *P. westermani*. Some eggs in the specimens were used for DNA extraction and molecular identification.

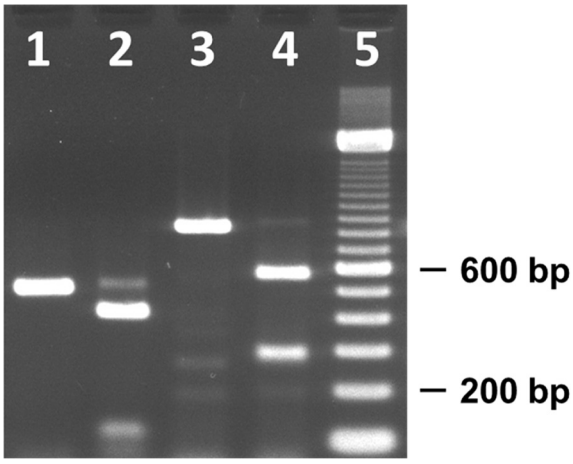

Fig. 4. RFLP patterns of PCR products amplified from the DNA of *Paragonimus* eggs from the patient. The ITS2 region of the nuclear ribosomal DNA was amplified by PCR and treated with endonuclease *Bss*SI, the enzyme for *P. miyazakii* (lane 1) or *Sna*BI, the enzyme for *P. westermani*, (lane 2). The mitochondrial 16S ribosomal RNA gene was amplified and treated with endonuclease *Sna*BI, the enzyme for diploid *P. westermani*, (lane 3) or *Bsr*DI, the enzyme for triploid *P. westermani*, (lane 4). A 100-bp DNA ladder marker was used to estimate the size of the fragments (lane 5). Based on the PCR-RFLP patterns, we identified the eggs as a triploid form of *P. westermani*.

3. Discussion

Human paragonimiasis is a food-borne zoonosis that results from the ingestion of raw or undercooked freshwater crustaceans or wild boar meat infected with metacercariae. Human paragonimiasis requires 2 intermediate hosts: a snail, where the embryonated eggs develop into cercariae, and a freshwater crustacean (e.g., crab or crayfish), where they develop into metacercariae. Metacercariae are passed to the definitive host (human being or carnivorous mammal) when the crustaceans are ingested in an undercooked state [3]. In Japan, *P. westermani* and *P. miyazakii* are common pathogens, and the majority of human paragonimiasis cases are caused by *P. westermani* [1]. Among Japanese men, human paragonimiasis occurs predominantly in middle- and old-aged individuals who consume wild boar meat or freshwater crab [2,3]. *P. westermani* exists in both diploid and triploid forms. The triploid form of *P. westermani* is presumed to be more pathogenic in humans than the diploid form [4]. The pathology of the conditions caused by the two forms differs; triploid flukes mainly form cysts in the lungs, whereas the diploid flukes cause lesions in the pleural cavity and pleura [5]. Previously, human paragonimiasis was endemic in Japan. However, re-emergent cases have been reported. In a case series of 443 patients with human paragonimiasis, Nagayasu et al. reported that the majority of the patients were residents of Kyushu Island, and immigrants (mostly from China, Thailand, and Korea) accounted for a quarter of the cases [3]. In this case, although he denied obvious consumption of raw or inadequately cooked freshwater crab, crayfish, wild boar, or deer meat, the time of infection was likely between 13 months earlier and 10 months earlier, judging by the timing of his eosinophilia

(Table 1). Furthermore, since he traveled to China during that same time, this could further support the infection time.

Patients with human paragonimiasis exhibit a wide variety of nonspecific findings on physical examination, chest radiographs, and CT scans. Therefore, human paragonimiasis may sometimes be misdiagnosed as pulmonary tuberculosis or lung cancer [6]. Pleuropulmonary paragonimiasis appears on radiographs as patchy airspace consolidation with or without cysts, ring shadows, subpleural linear opacities, and bilateral pleural effusions [7]. High-resolution CT findings consist of worm cysts, peripheral density, bronchial wall thickening, centrilobular nodules, masses, and infiltrative opacity [8]. Consequently, abnormal image findings often lead attending physicians to suspect malignant lung disease or lung tuberculosis. When these diseases are suspected, patients may be subjected to expensive and invasive examinations.

In our patient, an accurate diagnosis of *P. westermani* infection was made on discovering parasite eggs in his sputum, but the egg detection rate (in the bronchoscopic fluid, sputum, pleural fluid, stool) is very low at present, probably as a result of low-density infections [3,9]. Thus, immunodiagnosis may be the only reliable way to diagnose human paragonimiasis. Immunoblot tests reportedly have a sensitivity and specificity of 96 and 99 percent, respectively [10]. Unfortunately, immunoblot tests for the diagnosis of human paragonimiasis are not commercially available in Japan, so a multiple-dot ELISA for parasite-specific IgG antibodies against 12 different parasitic antigens is used. Although Nakamura et al. reported that the sensitivity for diagnosing human paragonimiasis is 90%, the specificity remains to be determined [9]. Moreover, a cross-reaction may occur with multiple-dot ELISA [10,11]. Therefore, when a patient's serum tests positive for an antigen in multiple-dot ELISA, the reactivity of the patient's serum to the suspected antigen needs to be confirmed by a double immunodiffusion test in agarose (Ouchterlony method) or combinations of binding and binding-inhibition ELISAs [12].

Morphological features of eggs such as shape, size, and shell character can be used to distinguish between *Paragonimus* species. However, the eggs of many Paragonimus species have overlapping morphological features that make species identification impractical. Therefore, the eggs were subjected to molecular identification by PCR-RFLP analysis, and they were identified as the triploid form of *P. westermani* (Fig. 3). The species and forms identified by the RFLP analyses were verified by sequencing the respective PCR products [13]. This allowed us to confirm that the ova were triploid forms of *P. westermani*.

In conclusion, a middle-aged man with a history of lung cancer who had undergone a lobectomy presented with *P. westermani* infection in a tertiary care cancer center. Even if patients deny having ingested raw or uncooked food, such as freshwater crab, wild boar, or deer meat, human paragonimiasis should be considered a possibility in the differential diagnosis for patients with increased serum eosinophils and chest radiological findings.

Conflict of interest

None.

References

[1] Mukae H, Taniguchi H, Matsumoto N, Iiboshi H, Ashitani J, Matsukura S, et al. Clinicoradiologic features of pleuropulmonary *Paragonimus westermani* on Kyusyu Island, Japan. Chest 2001;120:514–20.
[2] Uchiyama F, Morimoto Y, Nawa Y. Re-emergence of paragonimiasis in Kyushu, Japan. Southeast Asian J Trop Med Public Health 1999;30:686–91.
[3] Nagayasu E, Yoshida A, Hombu A, Horii Y, Maruyama H. Paragonimiasis in Japan: a twelve-year retrospective case review (2001-2012). Intern Med 2015;54:179–86.
[4] Blair D, Agatsuma T, Watanobe T, Okamoto M, Ito A. Geographical genetic structure within the human lung fluke, *Paragonimus westermani*, detected from DNA sequences. Parasitology 1997;115:411–7.
[5] Park GM, Im KI, Yong TS. Phylogenetic relationship of ribosomal ITS2 and mitochondrial COI among diploid and triploid *Paragonimus westermani* isolates. Korean J Parasitol 2003;41:47–55.
[6] Luo J, Wang MY, Liu D, Zhu H, Yang S, Liang BM, et al. Pulmonary paragonimiasis mimicking tuberculous pleuritis: a case report. Med (Baltimore) 2016;95:e3436.
[7] Im JG, Whang HY, Kim WS, Han MC, Shim YS, Cho SY. Pleuropulmonary paragonimiasis: radiologic findings in 71 patients. AJR Am J Roentgenol 1992;159:39–43.
[8] Akaba T, Takeyama K, Toriyama M, Kubo A, Mizobuchi R, Yamada T, et al. Pulmonary paragonimiasis: the detection of a worm migration track as a diagnostic clue for uncertain eosinophilic pleural effusion. Intern Med 2016;55:503–6.
[9] Nakamura-Uchiyama F, Onah DN, Nawa Y. Clinical features of paragonimiasis cases recently found in Japan: parasite-specific immunoglobulin M and G antibody classes. Clin Infect Dis 2001;32:e151–3.
[10] Slemenda SB, Maddison SE, Jong EC, Moore DD. Diagnosis of paragonimiasis by immunoblot. Am J Trop Med Hyg 1988;39:469–71.
[11] Tong DS, Liu YX, Tang F. Detection of four parasitic infections using ELISA. Zhongguo Xue Xi Chong Bing Fang Zhi Za Zhi 2013;25:327–8.
[12] Hoshina T, Tamura K, Kawano S, Kato T, Sato F, Horino T, et al. Two cases of *Paragonimiasis westermani* in a Chinese family diagnosed with the Ouchterlony double diffusion method. Kansenshogaku Zasshi 2014;88:866–70.
[13] Sugiyama H, Umehara A, Morishima Y, Yamasaki H, Kawanaka M. Detection of *Paragonimus metacercariae* in the Japanese freshwater crab, *Geothelphusa dehaani*, bought at retail fish markets in Japan. Jpn J Infect Dis 2009;62:324–5.