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Leptospirosis in the Tohoku Region: Re-emerging Infectious Disease

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Leptospirosis is a zoonotic and disaster-related infectious disease. It is mainly endemic in subtropical or tropical countries and has not been reported since 2009 in the Tohoku region (northern Japan), including the Yamagata and Miyagi Prefectures. However, we experienced four patients with leptospirosis in the Tohoku region from 2012 to 2014; three patients (#1-3) live in the agricultural areas of the Yamagata Prefecture and one patient (#4) was a visitor to the Miyagi Prefecture. Patient 1 (81-year-old female) is a villager, with a rat bite, while Patient 2 (77-year-old male) and Patient 3 (84-year-old female) are farmers and were infected probably during agriculture work. Patient 4 (40-year-old male US citizen) was infected while traveling in Thailand. They had chief complaint of fever, headache, and myalgia and showed manifestations of hyperbilirubinemia (mean, 4.35 mg/dL), thrombocytopenia and acute kidney injury (AKI). All patients were diagnosed by polymerase chain reaction using blood and/or urine samples and a microscopic agglutination test for the anti-*Leptospira* antibody. All the patients were treated with infused antibiotics, including minocycline. The patients underwent hemodialysis due to severe AKI (mean serum creatinine, 4.44 mg/dL), except for Patient 2 with the normal serum creatinine level (1.12 mg/dL). All the patients recovered and were discharged. The presence of the three patients in the Yamagata Prefecture implies that leptospirosis does re-emerge in the Tohoku region. Therefore, careful survey of the pathogen is necessary for febrile patients with AKI who engage in agriculture or have a recent history of travelling in subtropical or tropical countries.

Keywords: disaster; global warming; hemodialysis; leptospirosis; outbreak
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Introduction

Leptospirosis is a zoonotic disease that can spread worldwide, and this endemic mainly occurs in countries with humid subtropical or tropical climates; thus, it has the potential to become an epidemic because leptospirosis is a percutaneous or transmucosal infection due to increased contact with animal urine-contaminated water (World Health Organization 2003). The International Leptospirosis Society estimates that 300,000-500,000 cases of leptospirosis occur annually worldwide (World Health Organization 1999; Hartskeerl 2006). A total of 5-10% of patients with leptospirosis can potentially develop a severe form of the disease, with a fatality rate of > 10% for Weil's disease and

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up to 70% for leptospirosis pulmonary hemorrhage syndrome (Bharti et al. 2003; Dolhnikoff et al. 2007; Gouveia et al. 2008).

Annually, 20-30 patients with leptospirosis are reported in Japan, and about half of the cases live in the Okinawa Prefecture (Koizumi and Watanabe 2006). According to the national surveillance of leptospirosis in the Tohoku region from 2007 to 2011, only one patient was reported in the Akita Prefecture in 2008; therefore, leptospirosis is a very rare infectious disease in northern Japan (National Epidemiological Surveillance of Infectious Diseases (NESID)). Recently, people can suffer from leptospirosis more frequently in disaster such as flood, and leptospirosis is categorized as a disaster-related infectious disease (World Health Organization 2006; Amilasan et al. 2012). A cluster of leptospirosis, affecting three electrical specialists who ineffectively protected their legs from the mud, was reported in the Kochi Prefecture in 2011 (Infectious Agents Surveillance Report 2012). Another case in 2013 was an athlete in a triathlon who swam in a river that had become muddy due to a typhoon, and he was infected with *Leptospira* in the Shizuoka Prefecture (Infectious Agents Surveillance Report 2014).

Because of global warming, extreme weather events such as typhoons and floods are expected to occur with increasing frequency and a greater intensity. This may potentially result in an upsurge in the disease incidence even in northern Japan.

We treated four patients with leptospirosis in the Tohoku region (northern Japan) from 2012 to 2014, and we discuss the possibility of reemergence of the *Leptospira* infection.

Methods

Patients

Patients with leptospirosis were diagnosed and treated at the Okitama Public General Hospital, Shinjo Tokushukai Hospital, and Yamagata Prefectural Central Hospital in the Yamagata Prefecture and at the Tohoku University Hospital in the Miyagi Prefecture in Japan from 2012 to 2014.

Polymerase chain reaction and the microscopic agglutination test for diagnosing leptospirosis

Laboratory diagnosis of leptospirosis was performed by DNA detection and/or antibody detection. We performed *flaB*-nested polymerase chain reaction (PCR) to detect DNA in the plasma, serum, and/or urine followed by direct nucleotide sequencing of the amplicons (Koizumi et al. 2013). The anti-*Leptospira* antibody was detected by the microscopic agglutination test (MAT) using patients' serum samples with a battery of serovar strains, as described previously (Koizumi et al. 2008). At least a four-fold increase in the antibody titers between the acute and convalescent serum samples was judged as positive.

Clinical courses of the four patients: A summary of the four patients with leptospirosis encountered in the Tohoku region is presented in Table 1. Three patients in the Yamagata Prefecture included

Table 1. Summary of patients with confirmed leptospirosis.

Patient	Age/Sex	Occupation	Places of onset in Japan	Exposure	Chief complaint	Co-morbidity	Laboratory diagnosis	Antibiotics	Hemodialysis	Outcome
1	81/F	none	Yamagata Pref.	rat bite	appetite loss	hypertension	PCR (blood/urine), MAT	TAZ/PIPC MINO CTRX	yes	recovered
2	77/M	farmer	Yamagata Pref.	agricultural work	fever, myalgia	atrial fibrillation, gastric ulcer	PCR (urine), MAT	CMZ IPM/CS MINO	no	recovered
3	84/F	farmer	Yamagata Pref.	agricultural work	fever, arthralgia, malaise	none	PCR (urine), MAT	MEPM MINO PIPC CTRX	yes	recovered
4	40/M	US military	Miyagi Pref.	swimming in a river	fever, headache, abdominal pain	none	MAT	CFPM MINO	yes	recoverd

MAT, microscopic agglutination test; PCR, polymerase chain reaction; TAZ/PIPC, tazobactam/piperacillin; MINO, minocycline; CTRX, ceftriaxone; CMZ, cefmetazole; IPM/CS, imipenem/cilastatin; MEPM, meropenem; CFPM, cefepime.

Fig. 1. A rat bite in Patient 1.
The patient had a small rat bite on the fourth finger of the left hand.

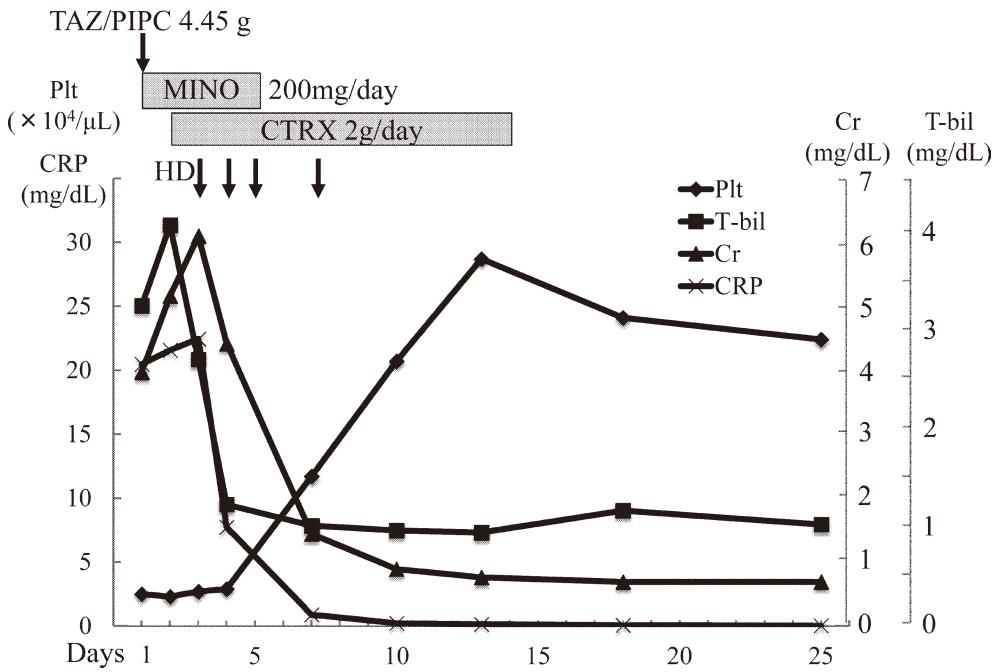

Fig. 2. The clinical course of Patient 1.
The patient sustained a rat bite on the finger in the middle of October in 2012 and presented with appetite loss with renal failure, icterus, and a severe decrease in the platelet count within a week. Leptospirosis was strongly suspected; thus, minocycline (MINO) and ceftriaxone (CTRX) were administered, and hemodialysis (HD) was performed four times. The symptoms of icterus, renal failure, and the low platelet count were recovered soon after treatment. Leptospirosis was confirmed by polymerase chain reaction on admission. TAZ/PIPC, tazobactam/piperacillin.

elderly farmers and villagers (mean age, 80.6 years), and there was another imported case in which a 40-year-old patient was infected while traveling in Thailand. We observed a wound on the finger due to a rat bite in Patient 1 (Fig. 1) and the subsequent onset of leptospirosis (Fig. 2). On the other hand, we speculated that Patients 2 and 3 had contacted leptospira organisms while performing agricultural work. The patient 4 swam in a river of Thailand and returned to the United States military base in Japan, and the patient had also been visiting the Japan Ground Self-defense Force camp in the Miyagi Prefecture. The initial chief complaints in the four patients were fever, loss of appetite, malaise, myalgia, arthralgia, headache, and abdominal pain, as previously reported (Amilasan et al. 2012). Laboratory findings showed severe AKI and a mild liver injury with hyperbilirubinemia (Table 2). A laboratory diagnosis of leptospirosis was confirmed by *flaB*-nested PCR from blood and urine samples and by MAT. Leptospiral *flaB* was detected in 3 patients; two in urine samples only and one in both blood and urine samples (Table 1). Based on the *flaB* sequences, all the *Leptospira* species were *L. interrogans* (data not shown). All four patients were serologically confirmed to have leptospirosis using paired serum samples (Table 3). The highest reactive serogroup(s) was Canicola (Patient 1), Autumnalis, Icterohaemorrhagiae and Javanica (Patient 2), and

Table 2. Summary of blood test data on admission.

	Patient 1	Patient 2	Patient 3	Patient 4
White blood cell (/μL)	12,600	9,500	15,760	6,000
Platelet (/μL)	25,000	23,000	22,000	109,000
T-bil (mg/dL)	3.2	7.1	5.5	1.6
AST (U/L)	113	186	31	48
ALT (U/L)	104	104	51	46
BUN (mg/dL)	56.4	21.5	91.5	39
Cr (mg/dL)	3.97	1.12	4.49	4.86
CRP (mg/dL)	20.47	24.66	18.60	29.00

Table 3. The results of microscopic agglutination test (MAT).

Leptospira			Patient 1 antibody titer		Patient 2 antibody titer		Patient 3 antibody titer		Patient 4 antibody titer	
Species	Serogroup	Serovar	Oct. 27, 2012	Nov. 9, 2012	Aug. 24, 2014	Sept. 10, 2014	Jul 23, 2013	Aug.19, 2013	Aug. 6, 2013	Aug. 19, 2013
borgpetersenii	Ballum	Castellonis	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160
borgpetersenii	Javanica	Javanica	< 40	< 160	< 40	320	< 40	< 160	< 40	640
borgpetersenii	Javanica	Poi	< 40	160	**< 40**	**1,280**	< 40	< 160	< 40	640
interrogans	Australis	Australis	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160
interrogans	Autumnalis	Autumnalis	< 40	< 160	< 40	320	< 40	640	< 40	< 160
interrogans	Bataviae	Bataviae	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160
interrogans	Canicola	Canicola	**< 40**	**2,560**	< 40	< 160	< 40	< 160	< 40	< 160
interrogans	Icterohaemorrhagiae	Copehhageni	< 40	1,280	< 40	160	**< 40**	**1,280**	< 40	< 160
interrogans	Hebdomadis	Hebdomadis	< 40	< 160	< 40	320	< 40	< 160	< 40	320
interrogans	Icterohaemorrhagiae	Icterohaemorrhagiae	< 40	1,280	**< 40**	**1,280**	**< 40**	**1,280**	**< 40**	**1,280**
interrogans	Hebdomadis	Kremastos	< 40	< 160	< 40	320	< 40	< 160	< 40	320
interrogans	Pomona	Pomona	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160
interrogans	Pyrogenes	Pyrogenes	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160
interrogans	Autumnalis	Rachmati	< 40	< 160	**< 40**	**1,280**	< 40	640	< 40	< 160
kirschneri	Grippotyphosa	Grippotyphosa	< 40	< 160	< 40	< 160	< 40	< 160	< 40	< 160

Icterohaemorrhagiae (Patients 3 and 4).

All patients were treated with infused antibiotics, minocycline (MINO) and ceftriaxone (CTRX), cefepime (CEPM) or imipenem/cilastatin (IPM/CS), and hemodialysis was needed for Patients 1, 3 and 4 due to severe AKI (Table 1), and they fully recovered. Fig. 2 shows the clinical course for Patient 1.

Discussion

Leptospirosis has been a very rare infectious disease in the Tohoku region since the 1980s (Akiyama et al. 2001), but we experienced three patients and one imported case from 2012 to 2014.

The three patients from the Yamagata Prefecture were old (mean age, 80.6 years), and two of them were female. These findings were quite different from the patients in the Philippines where 95% of the leptospirosis patients were male, and the median age was 30 years with a high mortality (Chagan-Yasutan et al. 2013). In Yamagata's agricultural areas, old women are the one of the main workers in the rice fields and the difference from Manila indicate that the victims of leptospirosis are those who can be easily exposed to contaminated water.

Leptospirosis was suspected in all of the patients, MINO was administered during the early stage of admission, and the diagnosis of leptospirosis was confirmed by PCR using blood and/or urine samples and by MAT. In addition, hemodialysis may be the most important factor for survival in these patients. It is necessary to promptly start life-saving hemodialysis if the patient with leptospirosis has an oliguric AKI (Daher et al. 2009).

Before the 1970s, leptospirosis was a common infectious disease in the Tohoku region, and during the outbreak of 1959, > 800 patients were observed in the Miyagi Prefecture; subsequently the number of patients dramatically decreased because of the increased use of farm machinery (Akiyama et al. 2001). Recently, outbreak of

leptospirosis after a flood was reported in Metro Manila, the Philippines in 2009 (Amilasan et al. 2012), and unpredictable severe rainfall during the summer was observed in all areas over Japan. For example, in the Yamagata Prefecture, localized torrential downpours during rainy season in July 2013 created landslides, muddy rivers, and floods over large areas. Typhoon No. 8 made landfall on July 9, 2014, causing severe flooding in the rice fields of Nanyo City, the Yamagata Prefecture. These localized torrential downpours with floods and muddy waters may have created the potential for leptospiral infections. Since Patients 2 and 3 did not have apparent exposure to muddy water in such disasters, we assume that they were infected with *Leptospira* during their regular agricultural work, which has increased contact with animal urine-contaminated water. However, these cases indicate that *Leptospires* live in the environment of rural area and there is the potential for an epidemic of leptospirosis after a flood in the Tohoku region.

The DNA of the pathogenic *Leptospira* was confirmed in the soil of the coastal area in the Philippines after a storm surge, indicating that the soil can be a breeding ground for *Leptospira* during a disaster and the agents can survive in seawater (Saito et al. 2014). Although historical huge tsunami hit the east coastline of the Tohoku region in March 11, 2011, the patients of leptospirosis were not found (National Epidemiological Surveillance of Infectious Diseases (NESID)). However, our findings here imply that careful evaluation of pathogens in febrile patients associated with AKI after disaster including Tsunami would be necessary.

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Conflict of Interest

The authors declare no conflict of interest.

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