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Abstract

Japanese spotted fever (JSF) is a tick-borne rickettsial disease prevalent in western Japan, with an increasing incidence and geographical distribution. This retrospective study aimed to describe the clinical features of JSF and identify factors associated with its severe form. We included adult patients with laboratory confirmed JSF in Nagasaki prefecture from 2010 to 2021. Severe JSF was defined by the presence of altered mental status, low blood pressure, or low oxygen saturation. In total, 65 JSF cases were diagnosed. Common symptoms were fever (87%), rash (48%), and fatigue (48%) with eschars detected in 50 (79.4%) of patients. Thirty-eight (60.3%) patients were initially diagnosed with non-JSF conditions. Twenty-one (33.3%) cases were categorized as severe JSF, including one death. Pre-hospital factors associated with severe JSF included age ≥75 (adjusted odds ratio [aOR] 37.53, 95% confidence interval [CI] 3.03-465.38), male sex (aOR 26.5, 95% CI 4.23-166.00), and a treatment delay ≥4 days from onset (aOR 5.96, 95% CI 1.13-31.60). This study highlights diagnostic challenges of JSF due to its non-organ-specific clinical presentation. Delayed initial treatment, advanced age, and male sex significantly increase the risk of severity. It is crucial to further raise awareness of JSF among clinicians and residents in endemic areas.

Introduction

Japanese spotted fever (JSF) is a tick-borne rickettsial disease caused by *Rickettsia japonica*, primarily endemic in western Japan (1). Since its first report in 1984 (2), the number of JSF cases has gradually increased. In 2020, 422 cases were reported in Japan (3). The classic triad of JSF is fever, an erythematous rash, and eschar (4). However, clinical diagnosis is challenging due to non-specific symptoms and the difficulty in identifying the characteristic eschar, especially for clinicians inexperienced with the disease. In a study of JSF in Hiroshima prefecture, one-third of patients were misdiagnosed at the first medical presentation (5). A study in 2018 revealed that a variety of infectious and non-infectious diseases was initially suspected in cases later confirmed as rickettsioses (6).

The impact of JSF has grown with the expansion of its endemic region. Surveillance data from the National Institute of Infectious Diseases (NIID) showed that the number of prefectures where JSF cases were reported was 24 by 2005, which increased to 40 by 2019. An increasing number of cases has been reported in eastern Japan, an area not previously considered endemic (7). This is hypothesized to be attributable to climate change and growing populations of wild reservoir animals such as deer and boar (8,9) although increased disease awareness among local clinicians may also play a role. JSF

has recently been reported in other East Asian countries, including South Korea and China (10,11). To improve the accurate and timely diagnosis and treatment of JSF, clinicians across this region need to become more familiar with its clinical and epidemiological features.

The mortality rate of JSF is not insignificant, ranging from 1.1% to 4.1% (7). However, the risk factors of severe JSF are not yet well established, partly because the number of severe cases in studies to date has been insufficient for multivariable analysis. The definition of severe JSF also varies across studies, in which death, disseminated intravascular coagulation (DIC) or requirement for hemodialysis have each been reported as severe outcomes (5,12-15).

This study aims to describe and analyze the clinical characteristics of JSF and identify risk factors for severe disease.

Materials and Methods

Study design and setting

The study employed a multicenter, retrospective, and observational design, conducted in Nagasaki Prefecture, which reports approximately 10 to 20 JSF cases annually. The study sites encompassed four diverse healthcare facilities: Nagasaki University Hospital

(NUH), a tertiary referral center with 874 beds, located in Nagasaki city, the capital of Nagasaki prefecture; Nagasaki Rosai Hospital (NRH), a 350-bed secondary care hospital in Sasebo city, which is the second largest city of the prefecture; Nagasaki Medical Center (NMC), a tertiary hospital with 643 beds in Omura city, which is the fourth largest city; and Nagasaki Kamigoto Hospital (NKH), the sole, 186-bed general hospital on the remote Kamigoto island. The geographical location of Nagasaki prefecture and the respective locations of the four study sites are illustrated in Figure 1. For the identification of cases, a comprehensive search of hospital medical records was undertaken, spanning from January 2010 to December 2021.

Diagnosis and definition

The study included patients aged 18 years or older with a laboratory-confirmed diagnosis of JSF. Laboratory testing of samples from patients residing in Nagasaki city was conducted at the Nagasaki-shi-Hokenkankyo-shikenjo (Nagasaki City Health and Environmental Test Center), and samples from patients dwelling outside Nagasaki City were tested by the Nagasaki Prefectural Environment and Health Research Center. Laboratory confirmation of JSF was established through either nested polymerase chain reaction (PCR) or serological antibody testing. The nested PCR process involved the

analysis of patient blood or eschar samples and targeted the gene encoding the 17-kDa protein, a ubiquitous antigen among *Rickettsia* species. Serum antibody titers were measured by an indirect immunofluorescence assay, noted for its extensive cross-reactivity within the spotted fever group of rickettsial diseases. The assay was considered positive when IgM or IgG titers, measured during acute and convalescent phases, showed an increase greater than four-fold, or a single acute serum IgM-titer was more than 80. In routine clinical practice, blood samples from every suspected case were forwarded to local public health reference laboratories for PCR analysis, and eschar samples were tested if recognized by a clinician. Serum samples were subjected to paired serum antibody testing primarily when PCR results were negative. In the current study, five cases were exceptionally tested for serum antibodies subsequent to receiving a positive PCR result.

Patients were classified as having severe JSF if they met at least one of the following criteria during their clinical course: a Glasgow Coma Scale less than 15, an oxygen saturation (SpO_2) in ambient air below 94% or requiring oxygen supplementation, or a systolic blood pressure lower than 90 mmHg or requiring vasopressors.

Data collection

For each patient, the following information was collected: age, sex, height, weight, postal address, date of onset and diagnosis, history of outdoor activities, underlying diseases, presenting clinical features, laboratory findings, treatment regimen, clinical course and outcome. Onset was defined as the date on which JSF-related symptoms such as fever, fatigue, myalgia, appetite loss or headache were recognized by patients. Fever was defined as a body temperature of 37.5°C or greater. Outdoor activities included forestry work, farming, gardening, and other activities that pose a risk of tick exposure. Cardiovascular diseases included conditions that affect cardiac function, such as coronary artery disease, congestive heart failure, or cardiomyopathy. Chronic pulmonary diseases were defined as any chronic condition that affects respiratory function, such as chronic obstructive pulmonary disease, bronchial asthma, or bronchiectasis. Active cancer was defined when patients had a confirmed diagnosis of any type of cancer with either of the following: patients had not undergone any curative treatment; or patients had undergone any curative treatment within the past 6 months of the diagnosis of JSF. The date of diagnosis was the date on which a clinician identified rickettsiosis as a probable diagnosis and initiated treatment. Treatment for JSF was

defined by the administration of either oral or intravenous tetracycline. All patients were followed up until the end of the treatment; there was no loss to follow up.

To assess the representativeness of the data from the study sites, we obtained demographic data of JSF cases in Nagasaki prefecture published online by NIID (3).

Statistical analysis

To test for differences in baseline characteristics between severe and non-severe cases, Fisher's exact test was applied for categorical and Mann-Whitney U test for continuous variables. The estimation of odds ratios (OR) and 95% confidence intervals (CI) pertinent to severe disease was conducted through logistic regression analysis. The multivariate regression analysis for severe JSF included the following pre-determined variables: age, sex, the presence of underlying disease, the interval from onset to initiation of treatment, and body mass index (BMI). Previous studies have reported that severe JSF patients tend to be older and male, but these associations are mostly non-statistical due to the small sample sizes (Table 1) (5, 12-15). Higher age and the presence of underlying disease have been reported to be associated with severe Mediterranean spotted fever (MSF) (16), while male sex and treatment delay have been linked to severe Rocky Mountain spotted fever (RMSF) (17,18). BMI was also included

in the model since certain infectious disease outcomes are associated with obesity (19). A 2-sided *P*-value of less than 0.05 was considered indicative of statistical significance for all tests. Statistical analysis was performed with Stata, version 17.0 (StataCorp LLC, College Station, Texas, USA).

Ethics approval and consent to participate

This research was conducted in compliance with the Declaration of Helsinki and the Ethical Guidelines for Medical Research Involving Human Subject issued by the Ministry of Education, Culture, Sports, Science, and Technology. This research was approved by Nagasaki University Hospital Clinical Research Ethics Committee (Approval number: 22062021). We applied the opt-out approach to obtain consent from participants. The Nagasaki University Hospital Clinical Research Ethics Committee waived the need for specific informed consent for this retrospective, non-invasive study.

Results

Baseline, admission and clinical characteristics of JSF cases

From January 2010 to December 2021, 65 JSF cases were confirmed at four hospitals. There was one neonatal case and one duplicate case referred from NKH to NMC thus

63 JSF cases were included for analysis. In total, 21 (33.3%) were categorized as severe JSF. Three cases required intensive care treatment, including one 58-year-old male patient without any comorbidities who died after 2 days of hospitalization, resulting in the case fatality rate of 1.6% (95%CI 1.4-7.6). Patients are suspected to have contracted the infection in Nagasaki Prefecture in all cases, except for one instance where the patient had a history of outdoor activity in Saga, a neighboring prefecture. None of the patients had a prior history of international travel.

There was clear seasonality of JSF cases, with none reported during the winter period from November to February (Figure 2a). The annual number of reported cases showed an increasing trend over the decade (Figure 2b). No statistically significant difference in severity was shown according to the reported month or year.

The overall characteristics of patients upon hospital presentation are shown in Table 2. Twenty-eight (55.6%) patients were male, though patients with severe JSF tended to be male ($p<0.01$). The median (IQR) age of patients was 72 years (64-80.5), and this was not statistically different according to severity. Fever was the most frequent complaint followed by rash and fatigue. Skin rash was reported by less than half of patients but was identified by clinical examination in all cases. Fifty-five eschars were recognized in 50 patients (79.4%), of which 24 (43.6%) were detected in the lower extremities, 16

(29.1%) in the trunk, 10 (18.2%) in the upper extremities, and 5 (9.1%) in the head, face, or neck.

Initial diagnosis and laboratory findings

Thirty-eight (60.3%) patients were not diagnosed with JSF at the first medical attention. Ineffective antibiotics against JSF, including cephalosporins, carbapenems, or macrolides were administered to those considered to have bacterial infections other than rickettsiosis. For those who were not diagnosed with JSF at first presentation, the median time from presentation to treatment for JSF was 3 days (IQR 2-4).

Common laboratory abnormalities, each demonstrated in at least 80% of patients, were: elevated C-reactive protein; elevated D-dimer; elevated aspartate aminotransferase (AST) to alanine aminotransferase (ALT) ratio; elevated AST; elevated lactate dehydrogenase (LDH); hyponatremia; thrombocytopenia, and positive urine protein.

Forty-seven (74.6%) patients were positive by eschar or blood PCR. The positive rate of eschar PCR, blood PCR, and serum antibody tests were 86.7% (39/45), 31.1% (14/45), and 100% (21/21), respectively. The positive rate of blood PCR was higher in severe cases compared to non-severe cases (56.3% vs 17.2%, p=0.016).

Treatment and outcome

Fifty-eight cases (92.1%) were hospitalized, with a median (IQR) duration of 12 (9-15) days and ranging from 2 to 83 days. The median (IQR) days from disease onset to the initiation of effective treatment was 4 (3-5) days, ranging from 0 to 12 days. Patients with severe JSF tended to have a treatment delay of four days or more (71.4% vs 48.8%, p=0.09).

Fifty-two (82.5%) cases were treated with tetracycline alone. Patients with severe JSF tended to receive dual antibiotic therapy with tetracycline and quinolone (28.6% vs 9.5%, p=0.08). One non-severe case spontaneously improved without treatment. The median (IQR) duration of antibiotic treatment was 12 (9-14) days, and this did not differ statistically according to severity.

Factors related to severe JSF

Pre-hospital characteristics were further scrutinized by multivariate analysis (Table 3). Whereas male sex was the only factor associated with severe JSF in univariate analysis, in this multivariate model, beside male sex (adjusted OR [aOR] 26.5, 95% CI 4.23-166.0), an age of 75 years or older (aOR 37.53, 95% CI 3.03-465.38) and an interval from onset to treatment of four days or more (aOR 5.96, 95% CI 1.13-31.60)

were significantly associated with severe JSF. As a sensitivity analysis, DIC defined by Japanese Association for Acute Medicine (20) was added to our severity criteria. When including DIC among the severe cases, the same risk factors were identified in multivariate analysis except for delay in treatment which showed no statistical association.

Discussion

Unlike previous, mostly single-centered studies of JSF, in this study cases were recruited from four sites comprising two tertiary hospitals, a secondary hospital, and the sole community-hospital of a remote island. During the same study period, 154 cases were officially reported in Nagasaki prefecture including 90 (58.4%) patients aged 70 or older and 58 (37.7%) male patients. Hence, our study cohort is representative of cases across Nagasaki prefecture in the same period. Cases were reported from March to October, most frequently in September and October, and the increasing number of annual cases was consistent with the national trend (21).

The results that 25 (39.7%) in our study were diagnosed with JSF at the first medical attention highlights the diagnostic challenges of this disease. This can be partly

attributed to the non-specific clinical presentation of JSF with fever, rash, fatigue, headache, arthralgia or myalgia, with or without eschar (22). The rates of fever, erythematous rash, and eschar were similar to past reports (fever 74-100%, rash 86.6-100%, and eschar 61.8-89%, respectively) (5,6,12-15,22). Among those who were not diagnosed with JSF at the first medical presentation, bacterial infection was the most frequent diagnosis.

Despite these challenges, epidemiological knowledge and a careful clinical history can alert attending physicians to the possibility of JSF, as all patients in our study either resided in endemic areas or had a history of outdoor activities. Careful skin examination is necessary to detect rash and eschar, although the absence of skin lesions does not exclude the possibility of JSF. In addition, laboratory results may help clinicians to suspect JSF: low platelet count and sodium, high AST, LDH, and CRP, and abnormal urinalysis were common among our JSF cases, consistent with other studies (5,6). Additionally, an AST/ALT ratio >1 was one of the commonest laboratory abnormalities seen in our study, present in 58 (92.1%) cases. An elevated AST/ALT ratio has been reported in scrub typhus patients (23), which may indicate a common pathology among

rickettsial diseases. Further studies are required on the utility of AST/ALT ratio for diagnosing rickettsioses.

This was the first multivariable analysis demonstrating that advanced age, male sex, and delayed treatment were significantly associated with severe JSF. Although our definition of severe JSF, which encompasses blood pressure, respiratory condition, and conscious level differs from previous studies, we believe this vital signs-based definition accurately captures severe cases since these three components also constitute the quick Sequential Organ Failure Assessment score, which is the diagnostic criteria for sepsis in the early phase of disease (24).

In our study, respiratory failure was more frequent in male patients compared with female patients (OR 9.24, 95% CI 2.29-37.38, p<0.01) (Table 4). There is evidence for sex-based differences in immune responses to infection and clinical outcomes (25), though the specific immunopathogenesis of JSF remains to be determined. It is possible that smoking history, which we did not ascertain in this retrospective study, could have confounded this association between sex and respiratory failure: smoking history should be included in future risk factor analyses for severe JSF. A recent study utilizing a

national inpatient database and including 1,360 JSF patients showed that delayed initiation of tetracycline was associated with an increase in mortality (26). Nevertheless, in our study, out of eight patients whose interval between onset and initiation of treatment was within a day, five patients had severe JSF. Therefore, while treatment delay can contribute to disease severity in JSF, there are likely to be other important host and clinical determinants. Further studies with a suitably large sample size are needed to determine the relative contribution of treatment delay and other factors to the risk of severe JSF.

There is considerable evidence for the association between obesity and the severity of respiratory tract infections such as severe COVID-19 and numerous immunopathogenic mechanisms have been postulated (27). However, no previous study has reported on this association in rickettsial diseases. In our study, patients with severe JSF tended to have higher BMI than those with non-severe JSF but this association was not statistically significant even after adjusting for age and gender.

In this study, blood PCR was less frequently positive than eschar PCR, but blood PCR positivity was significantly associated with severe disease. To our knowledge, this is the

first study describing the association between blood PCR positivity and severity in rickettsiosis. It is notable that patients with severe disease complained of fever less frequently, and their median body temperature was lower than that in non-severe patients. This result provides validity to non-statistical differences reported in previous studies. Hatano et al reported that the mean body temperatures of severe and non-severe patients were 37.7°C and 38.5ºC, respectively (5). The median body temperatures in DIC and non-DIC patients were 37.6°C and 38.5°C, respectively, in a study by Miyashima et al (14). This finding may reflect differences in the immune responses of patients with severe and non-severe JSF, but further investigations are required to characterize and contrast the immune pathways involved.

There are some limitations in our study. First, since this is a hospital-based study, we might have been unable to include mild cases treated at clinics. Second, our diagnostic methods were not able to differentiate JSF from other spotted fever group rickettsiae found in Japan, such as *R. helvetica*, *R. heilongjiangensis*, or *R. tamurae*. Third, compared to previous studies, a substantial number of our study cases were included from the university hospital. Consequently, it is possible that our study selected for more severe cases. Nonetheless, this difference in the characteristics of study sites does not

entirely account for the high incidence of severe cases, as the highest percentage of severe cases was noted at NKH which is not a tertiary care hospital. Further studies are needed to elucidate the cause of high prevalence of severe JSF at NKH.

In conclusion, this study emphasizes the need to keep raising the awareness of clinicians regarding JSF, to improve prompt, accurate diagnosis and the timely initiation of treatment. Advice to avoid tick-bites for residents in endemic areas, especially elderly male patients who are at a higher risk of severe disease, should be regularly updated and distributed. Our study contributes to the growing evidence base for clinical and laboratory predictors of severe JSF: future large-scale, prospective studies, including varied sites, are needed to further characterize and explain these associations.

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Conflict of interest

The authors declare that they have no competing interests.

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Figure legends.

Figure 1. Location of Nagasaki prefecture and four study sites.

1, Nagasaki University Hospital. 2, Nagasaki Rosai Hospital. 3, Nagasaki Kamigoto Hospital. 4, Nagasaki Medical Center.

Figure 2. Trends in Japanese spotted fever (JSF) cases by month and year.

(A) Number of cases by month. (B) Number of cases by year. Dark gray columns show the number of severe JSF cases, and light gray columns show the number of non-severe JSF cases.

Table 1. Summary of previous studies analyzing risk factors for severe Japanese spotted fever.

Author, publication year	Severe case definition	Severe / Total cases	Age			Male sex, n (%)		
			Severe	Non-severe	*P* value	Severe	Non-severe	*P* value
Kodama, et al., 2003	1. DIC 2. Respiratory failure 3. Impaired consciousness	6/28	69[1)]	66.5	0.614[3)]	6 (100%)	17 (77.3%)	0.553[3)]
Nakamura, et al., 2011	1. In-hospital death 2. DIC	6/51	75.5[1)]	61.9	0.43	2 (33.3%)	22 (48.9%)	0.67
Miyashima, et al., 2018	1. DIC	5/20	68.8[2)]	68	0.45	4 (80%)	4 (27%)	**0.035**
Hatano, et al., 2021	1. Death 2. Require hemodialysis	6/82	76.2[1)]	73.9	0.618	5 (83.3%)	29 (38.2%)	0.077
Sakabe, et al., 2021	1. Death	8/239	76.7[1)]	69	0.19	No data		

DIC, disseminated intravascular coagulation. Bold text indicates statistical significance.

[1)] Mean age

[2)] Median age

[3)] *P* values calculated by us using Mann-Whitney U test (age) and Fisher's exact test (sex).

Table 2. Characteristics of 63 patients with confirmed Japanese spotted fever according to disease severity.

Characteristics	Total	Severe	Non-severe	OR (95% CI)	*P* value
Age, median years (IQR)	72 (64-80.5)	76 (70-82)	69.5 (61-80)		0.11
<65, n (%)	17/63 (27.0%)	3/21 (14.3%)	14/42 (33.3%)	Ref	Ref
65-74, n (%)	19/63 (30.2%)	7/21 (33.3%)	12/42 (28.6%)	2.72 (0.57-12.91)	0.21
≥75, n (%)	27/63 (42.9%)	11/21 (52.4%)	16/42 (38.1%)	3.21 (0.74-13.87)	0.12
Sex, n (%)					
Female	35/63 (44.4%)	5/21 (23.8%)	30/42 (71.4%)	Ref	Ref
Male	28/63 (55.6%)	16/21 (76.2%)	12/28 (28.6%)	8 (2.05-31.28)	<0.01
BMI, median (IQR)		23.1 (20.5-25.3)	21.9 (20.8-24.4)		0.61
<25 kg/m^2	43/63 (68.3%)	14/21 (66.7%)	29/42 (69.1%)	Ref	Ref
≥25 kg/m^2	12/63 (19.0%)	6/21 (28.6%)	6/42 (14.3%)	2.07 (0.57-7.59)	0.27
No data	8/63 (12.7%)	1/21 (4.8%)	7/42 (16.7%)	0.3 (0.03-2.64)	0.28
Study site					
Nagasaki University Hospital	32/63 (50.8%)	11/21 (52.4%)	21/42 (50.0%)	1.75 (0.40-7.69)	0.46
Nagasaki Kamigoto Hospital	13/63 (20.6%)	6/21 (28.6%)	7/42 (16.7%)	2.86 (0.53-15.47)	0.22
Nagasaki Rosai Hospital	13/63 (20.6%)	3/21 (14.3%)	10/42 (23.8%)	Ref	Ref
Nagasaki Medical Center	5/63 (7.9%)	1/21 (4.8%)	4/42 (9.5%)	0.83 (0.07-10.60)	0.89
Underlying diseases, n (%)					
Any	18/63 (28.6%)	7/21 (33.3%)	11/42 (26.2%)	1.41 (0.45-4.45)	0.56
Diabetes	9/63 (14.3%)	4/21 (19.0%)	5/42 (11.9%)	1.74 (0.41-7.45)	0.45
Cardiovascular diseases	5/63 (7.9%)	2/21 (9.5%)	3/42 (7.1%)	1.37 (0.21-9.04)	0.74
Chronic pulmonary diseases	5/63 (7.9%)	1/21 (4.8%)	4/42 (9.5%)	0.48 (0.05-4.66)	0.52
Active cancer	2/63 (3.2%)	1/21 (4.8%)	1/42 (2.4%)	2.05 (0.12-35.49)	0.62

Clinical complaints, n (%)					
Fever	55/63 (87.3%)	15/21 (71.4%)	40/42 (95.2%)	0.13 (0.02-0.78)	0.02
Rash	30/63 (47.6%)	9/21 (42.9%)	21/42 (50.0%)	0.75 (0.26-2.18)	0.59
Fatigue	30/63 (47.6%)	13/21 (61.9%)	17/42 (40.5%)	2.39 (0.79-7.22)	0.11
Arthralgia or myalgia	16/63 (25.4%)	6/21 (28.6%)	10/42 (23.8%)	1.28 (0.39-4.23)	0.68
Headache	16/63 (25.4%)	3/21(14.3%)	13/42 (31.0%)	0.37 (0.09-1.54)	0.16
Physical findings					
Body temperature, ºC, median (IQR)	38.1 (37.4-38.8)	37.5 (36.7-38.6)	38.2 (37.7-39.0)		0.04
Rash, n (%)	63/63 (100.0%)	21/21 (100.0%)	42/42 (100.0%)	NA	NA
Eschar, n (%)	50/63 (79.4%)	16/21 (76.2%)	34/42 (81.0%)	0.75 (0.21-2.70)	0.75
Altered mental status[1)], n (%)	13/63 (20.6%)	NA	NA	NA	NA
SpO_2 <94%[1)], n (%)	16/63 (25.4%)	NA	NA	NA	NA
SBP <90mmHg[1)], n (%)	8/63 (12.7%)	NA	NA	NA	NA
Interval from onset to treatment, n (%)					
<4 days	27/62 (43.5%)	6/21 (28.6%)	21/41 (51.2%)	Ref	Ref
≥4 days	35/62 (56.5%)	15/21 (71.4%)	20/41 (48.8%)	2.63 (0.82-8.41)	0.09
Diagnosis at first consultation, n (%)					
Rickettsial infection	25/63 (39.7%)	8/21 (38.1%)	17/42 (40.5%)	Ref	Ref
Bacterial infection	13/63 (20.1%)	5/21 (23.8%)	8/42 (19.0%)	1.33 (0.32-5.38)	0.69
Fever with unknown cause	7/63 (11.1%)	3/21 (14.3%)	4/42 (9.5%)	1.59 (0.29-8.87)	0.6
Heat-related illness	5/63 (7.9%)	3/21 (14.3%)	2/42 (4.8%)	3.19 (0.44-23.01)	0.25
Viral infection	6/63 (9.5%)	0/21 (0.0%)	6/42 (14.3%)	NA	NA
Others	7/63 (11.1%)	2/21 (9.5%)	5/42 (11.9%)	0.85 (0.13-5.37)	0.86
Diagnostic methods, n (%)					

PCR positive	47/63 (74.6%)	17/21 (81.0%)	30/42 (71.4%)	Ref	Ref
PCR negative and antibodies positive	5/63 (7.9%)	1/21 (4.8%)	4/42 (9.5%)	0.44 (0.05-4.27)	0.66
PCR not done and antibodies positive	11/63 (17.5%)	3/21 (14.3%)	8/42 (19.0%)	0.66 (0.15-2.83)	0.74
Positive rate of each test, n (%)					
Positive eschar PCR	39/45 (86.7%)	13/16 (81.3%)	26 /29 (89.7%)	0.5 (0.09-2.92)	0.65
Positive blood PCR	14/45 (31.1%)	9/16 (56.3%)	5 /29 (17.2%)	6.17 (1.34-28.36)	0.02
Positive serum antibodies	21/21 (100.0%)	6/6 (100.0%)	15/15 (100.0%)	NA	NA
Laboratory parameters, n (%)					
WBC >8,600/μL	16/63 (25.4%)	7 /21 (33.3%)	9/42 (21.4%)	1.83 (0.56-6.02)	0.36
Platelet <158×10^3/μL	51/63 (81.0%)	19/21 (90.5%)	32/42 (76.2%)	2.97 (0.57-15.59)	0.31
PT-INR >1.0	47/59 (79.7%)	19/21 (90.5%)	28/38 (73.7%)	3.39 (0.64-18.09)	0.18
APTT >34 second	23/58 (39.7%)	12/20 (60.0%)	11/38 (29.0%)	3.68 (1.11-12.26)	0.03
D-dimer >1.0 ng/mL	48/49 (98.0%)	21/21 (100.0%)	27/28 (96.4%)	NA	1
Sodium <138 mmol/L	52/63 (82.5%)	19/21 (90.5%)	33/42 (78.6%)	2.59 (0.49-13.68)	0.31
Creatinine >1.08 mg/dL	18/63 (28.6%)	12/21 (57.1%)	6/42 (14.3%)	8 (2.02-31.72)	<0.01
AST >30 U/L	55/63 (87.3%)	19/21 (90.5%)	36/42 (85.7%)	1.58 (0.29-8.77)	0.71
ALT >36 U/L	37/63 (58.7%)	13/21 (61.9%)	24/42 (57.1%)	1.22 (0.41-3.60)	0.79
AST>ALT	58/63 (92.1%)	21/21 (100.0%)	37/42 (88.1%)	NA	0.16
LDH >222 U/L	55/63 (87.3%)	19/21 (90.5%)	36/42 (85.7%)	1.58 (0.29-8.77)	0.71
Total bilirubin >1.5 U/L	5/61 (8.2%)	4/21 (19.1%)	1/40 (2.5%)	9.18 (0.85-99.44)	0.04
CPK >216 U/L	20/55 (36.4%)	12/21 (57.1%)	8/34 (23.5%)	4.33 (1.23-15.25)	0.02
CRP >0.14 mg/dL	63/63 (100.0%)	21/21 (100.0%)	42/42 (100.0%)	NA	NA
Positive urine protein	44/55 (80.0%)	18/21 (85.7%)	26/34 (76.5%)	1.85 (0.42-8.11)	0.5
Positive urine occult blood	43/55 (78.2%)	18/21 (85.7%)	25/34 (73.5%)	2.16 (0.50-9.38)	0.34

Treatment, n (%)					
Only tetracycline	52/63 (82.5%)	15/21 (71.4%)	37/42 (88.1%)	Ref	Ref
Tetracycline and quinolone	10/63 (15.9%)	6/21 (28.6%)	4/42 (9.5%)	3.7 (0.91-15)	0.07
No treatment	1/63 (1.6%)	0/21 (0.0%)	1/42 (2.4%)	NA	NA
Days to defervescence, median (IQR)	3 (3-4)	4 (3-5.5)	3 (3-4)		0.02
Duration of treatment, median (IQR)	12 (9-14)	13 (9-14)	11 (9-14)		0.16
Duration of hospitalization, median days (IQR)	11 (8-15)	15 (9-28)	10 (8-13)		<0.01

OR, odds ratio; CI, confidence interval; IQR, interquartile range; BMI, body mass index; SpO_2, oxygen saturation; SBP, systolic blood pressure; PCR, polymerase chain reaction; WBC, white blood cell; ALC, absolute lymphocyte count; PT-INR, prothrombin time international normalized ratio; APTT, activated partial thromboplastin time; AST, aspartate aminotransferase; ALT, alanine aminotransferase; LDH, lactate dehydrogenase; CPK, creatinine phosphokinase; CRP, C-reactive protein. Cut-off values other than ALC, PT-INR, APTT, D-dimer, and CRP were quoted from “Guidelines for Clinical Laboratory Test (Japanese Society of Laboratory Medicine 2021)”. Cut-off values of PT-INR, APTT, D-dimer, and CRP were taken from the values approved at Nagasaki University Hospital.

[1)] At any point during the clinical course of infection.

Table 3. Multivariate logistic regression analysis of risk factors for severe Japanese spotted fever.

		Adjusted OR	95% CI	*P* value
Age				
	<65	Ref	Ref	Ref
	65-74	6.77	0.83-55.39	0.08
	≥75	37.53	3.03-465.38	<0.01
Sex				
	Female	Ref	Ref	Ref
	Male	26.5	4.23-166.0	<0.01
Underlying diseases				
	None	Ref	Ref	Ref
	Any	0.86	0.18-4.12	0.85
BMI				
	<25kg/m^2	Ref	Ref	Ref
	≥25kg/m^2	5.38	0.74-39.06	0.1
	No data	1.42	0.10-21.15	0.8
Interval from onset to treatment				
	<4 days	Ref	Ref	Ref
	≥ 4 days	5.96	1.13-31.6	0.04

OR, odds ratio; CI, confidence interval; BMI, body mass index.

Table 4. Comparison of clinical and laboratory characteristics between male and female patients.

	Total	Female	Male	OR	95% CI	*P* value
Symptoms						
Fever	55	30 (85.7%)	25 (89.3%)	1.39	0.30-6.39	0.72
Rash	30	15 (42.9%)	15 (53.6%)	1.54	0.57-4.18	0.45
Fatigue	30	15 (42.9%)	15 (53.6%)	1.54	0.57-4.18	0.45
Arthralgia	16	7 (20.0%)	9 (32.1%)	1.89	0.60-5.96	0.38
Headache	16	9 (25.7%)	7 (25.0%)	0.96	0.31-3.02	1
Objective rash	63	35 (100%)	28 (100%)	N/A	N/A	N/A
Objective eschar	50	29 (82.9%)	21 (75.0%)	0.62	0.18-2.12	0.54
Severe features						
Low blood pressure	8	3 (8.6%)	5 (17.9%)	2.32	0.50-10.69	0.45
Respiratory failure	16	3 (8.6%)	13 (46.4%)	9.24	2.29-37.38	<0.01
Impaired consciousness	13	4 (11.4%)	9 (32.1%)	3.67	0.99-13.59	0.06
Laboratory features						
DIC	18	9 (27.3%)	9 (34.6%)	1.41	0.46-4.30	0.58
AKI	17	6 (17.1%)	11 (39.3%)	3.12	0.98-9.99	0.09

OR, odds ratio; CI, confidence interval; DIC, disseminated intravascular coagulation; AKI, acute kidney injury.


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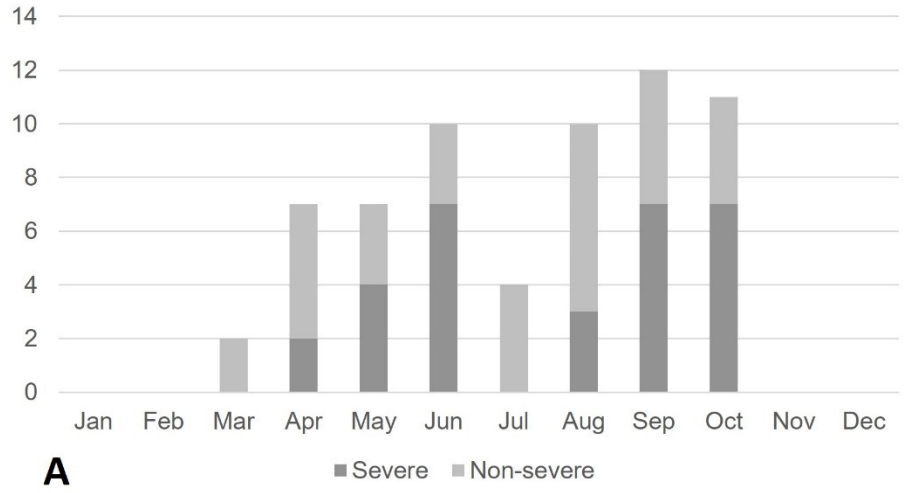
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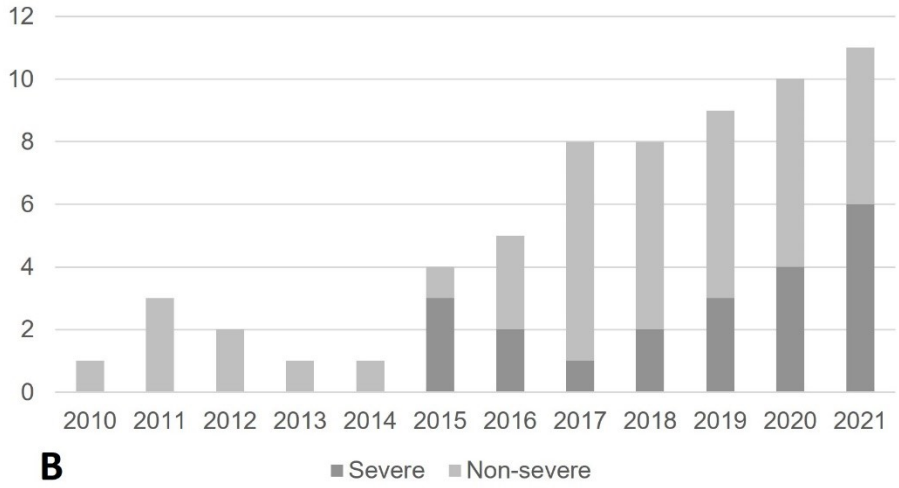
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