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BITING BY TICKS CARRYING RICKETTSIA TAMURAE IS NOT ALWAYS RISK FOR RICKETTSIA DISEASE IN AN IMMUNOCOMPETENT SUBJECT

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ABSTRACT
82 years old man was treated in the hospital for rhabdomyolysis and dehydration. Four days after admission, a bloated tick attaching to his arm was found. The result of PCR from his whole blood targeting rickettsia species including of JSF was negative. The serological study of anti *R.japonica* IgG/ IgM antibodies during both the acute and recovery phase did not appear to be the elevated levels. The removed tick was identical to that of *Amblyomma testudinarium* having *Rickettsia tamurae* from the result of PCR method. *R. tamurae* may be less infectious and virulent rickettsia compared *with R. japonica.*

INTRODUCTION

Rickettsia tamurae infection was firstly reported in 2010. The current case did not result in the infection in spite of being sucked by tick having *Rickettsia tamurae*. Here, we report not to be always at risk of being infected even if bitted by the tick with *Rickettsia tamurae.*

CASE

82 years old man with deterioration of consciousness was transported to the Shima Prefectural hospital by an ambulance. Clinical laboratory investigation revealed highly elevation of serum creatine phosphokinase (CPK) 14117 IU/l with high fever. He was diagnosed having rhabdomyolysis by dehydration. Four days after admission, a bloated tick attaching to his arm was found (Figure 1). No exanthema was detected on his whole body suggestive for the Rickettsia diseases. However, to rule out Japanese spotted fever (JSF), the polymerase chain reaction (PCR) examination for *Rickettsia japonica* (*R. japonica)* using peripheral blood and the serological study of anti *R. japonica* IgG/ IgM antibodies were performed at Mie Prefecture Health and Environment Research institute. The result of PCR from his whole blood by targeting rickettsia species including of JSF was negative. The serological study of anti *R.japonica* IgG/ IgM antibodies during both the acute and recovery phase did not appear to be elevated levels.

From the removed tick, a DNA sample was extracted with a QIAamp DNA mini kit (QIAGEN, Germantown, MD, USA), and was used for the tick specific PCR with the specific identification primers for tick species under the following conditions: one cycle of preheating (94C, 10sec), 30 cycles of denaturation (94C, 10sec), annealing (55C, 30 sec.), and extension (55C, 30sec), followed by one cycle of delay (72C, 5min) [1].

The sequence of the PCR product was identical to that of Simultaneously PCR using the primers targeting of the 17-kilo Dalton genus-common antigen gene of the rickettsia species was performed [2]. The sequence of the *Rickettsia* PCR product was identical to that of *Rickettsia tamurae (R. tamurae)*. Serological study for *R. japonica* in the patient's serum samples at the acute and recovered phase by the Mie Prefecture Health and Environment Research institute

Amblyomma testudinarium (A. testudinarium).
eliminated *R. japonica* infection. He developed no symptom of rickettsia infection during long term following up. We retried the recovery stage of serological study of anti *R. japonica* IgG after 7 month later on the first visit because he would have gotten enough level of anti *R.tamurae* IgG. But Anti Rickettsia antibody was not detected in his serum.

Figure 1. The swollen tick by sucking the patient's blood was shown. The tick attached to the patient skin at least more than four days.

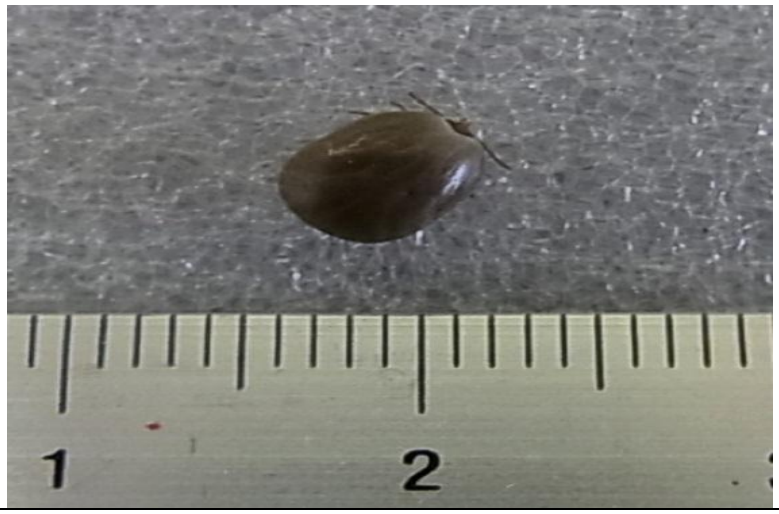

Table 1. The data of natural immune response is shown. The activity of natural killer cells and neutrophils seems to be normal. The inflammatory cytokine levels with stimulation were also within the normal range. E/T ratio: The number of effector cells and target cells in the mixture. Chromium release test to evaluate natural killer cell activity was performed using k-562 as target cell.

	Result	Unit	Average level
Natural killer cell active E/T ratio 10:1	19.7	%	8.9~29.5
Natural killer cell active E/T ratio 20:1	30.7	%	17.1~48.7
Phagocytosis by neutrophils	75.1	%	40.0~80.0
Bacteria-killing action by neutrophils	99.9	%	> 70.0 %
Serum IL-2 concentration	less than 5	pg/ml	less than 5
Serum IFN-γ concentration	less than 7.8	pg/ml	less than 7.8
TNF-α concentration	0.9	pg/ml	less than 2.8

DISUSSION AND CONCLUSION

R. tamurae infection was firstly reported in 2010 with the presence of the serological cross-reactivity between *R. tamurae* and *R. japonica* [3]. Based on DNA-based genetic testing and the size of the enlarged tick on his arm, the patient was obviously sucked by *A. testudinarium* for 4 days carrying *R. tamurae*. However, his serum samples at the acute and recovered phase had no immunoreactive or cross-reactive IgG to *R. japonica*. These data strongly suggest the failure of contagion after sucking of the blood by the tick carrying *R. tamurae*. A previously reported patient for *R. tamurae* infection had insulin-dependent diabetes mellitus, and was suspected to be non-fully immune-competent [3]. Therefore, we tested our patient's immune - competence including the natural immune activity following the Institutional Review Board (IRB) of Mie University Hospital approved protocol (Permit Number 2781). He showed no defects in the natural immune responses (Table 1).

The present data may suggest that *R. tamurae* was less infectious and virulent rickettsia compared *with R. japonica* for the immunocompetent subjects. This is only one case report and further study is required for declaration of the relations between *Rickettsia* infection and immune-competence of the patients.

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CONFLICT OF INTEREST

The authors have declared that no competing interests exist.

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