
Haruyuki Hirata, Satoru Kawai, Mari Maeda, Michio Jinmai, Kohei Fujisawa, Yuko Katakai, Kenji Hikosaka, Kazuyuki Tanabe, Yasuhiro Yasutomi, and Chiaki Ishihara*

School of Veterinary Medicine, Rakuno Gakuen University, Ebetsu, Japan; Laboratory of Tropical Medicine and Parasitology, Dokkyo Medical University, Tochigi, Japan; Corporation for Production and Research of Laboratory Primates, Hachimandai, Tsukuba, Japan; Laboratory of Malarialogy, International Research Center of Infectious Diseases, Research Institute for Microbial Diseases, Osaka University, Suita, Osaka, Japan; Laboratory of Immunoregulation and Vaccine Research, Tsukuba Primate Research Center, National Institute of Biomedical Innovation, Tsukuba, Ibaraki, Japan

Abstract. We demonstrate here the identification and phylogenetic characterization of Babesia microti (B. microti)-like parasite detected from a splenectomized Japanese macaque (Macaca fuscata fuscata) at a facility for laboratory animal science. On Day 133 after splenectomy, intra-erythrocytic parasites were found on light microscopic examination, and the level of parasitemia reached 0.3% on blood smear. Molecular characterization of the parasite using nested-polymerization chain reactions targeting the 18S rRNA, β-tubulin, and subunit 7 (eta) of the chaperonin-containing t-complex polypeptide 1 (CCT7) genes were identified as a B. microti-like parasite, designated the Japanese Macaque Babesia-1 (JM-1).

The genus Babesia belongs to the family Piroplasmida, closely related to Plasmodium and Theileria genera, and comprises over 70 species that parasitize mammals and birds. Babesia microti (B. microti) is a rodent-infective Babesia species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis. Nonhuman primates in species transmitted by ixodid ticks and is also a major etiologic agent of human babesiosis.
mixture contained 0.1 μg of template DNA, 5 μL of 10× PCR buffer with 15 mM MgCl₂ (TaKaRa Bio Inc., Shiga, Japan), 5 μL of dNTP mix (2 mM of each dNTP) (TaKaRa), 2.5 U of Takara LA Taq DNA polymerase (TaKaRa), and 50 pmol of each primer set for the 18S rRNA, β-tubulin, or CCT7 gene-specified primers for PCR as described previously with minor modifications as reported.21

NestPCR successfully amplified 18S rRNA, β-tubulin, and CCT7 genes from the gDNA of J79 (data not shown). The PCR products were isolated by 1.0% (w/v) agarose-gel electrophoresis in TAE buffer and purified with a GENECLEAN kit (BIO 101, Inc., Vista, CA). Nucleotide sequences of PCR products were isolated by 1.0% (w/v) agarose-gel electrophoresis in TAE buffer with 15 mM MgCl₂ (TaKaRa Bio Inc., Shiga, Japan), and 18S rRNA gene sequences were determined from a Hokkaido Squirrel.13 We refer to the cognate similarity, were from a rabbit. Although the origin of the JM-1 infection in the Japanese macaque is unclear, it is likely that the infection was from colonies of Japanese macaques at the facility of animal science laboratory or the cage outdoors at the breeding facility. Our case is most likely an example of subclinical or opportunistic infection that manifested in the postoperative period after splenectomy in a previously immunocompetent host.

### Table 1

<table>
<thead>
<tr>
<th>Target genes</th>
<th>Primers</th>
<th>Oligonucleotide sequences (5’ to 3’)</th>
</tr>
</thead>
<tbody>
<tr>
<td>18S rRNA</td>
<td>Piro 0F</td>
<td>GCCAGTAGTCTCATATGCTTTGTTTA</td>
</tr>
<tr>
<td></td>
<td>Piro 6R</td>
<td>CTCTTCTCTYTAAGTGATAAGTGTCAC</td>
</tr>
<tr>
<td></td>
<td>Piro 1F</td>
<td>CATGATGCTGCTCTWGTAYC</td>
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<tr>
<td></td>
<td>Piro 5.5R</td>
<td>CAGTTCAGTGTTAAGTTAC</td>
</tr>
<tr>
<td>β-tubulin</td>
<td>TUBU-ATG5F</td>
<td>ATGAGGAGARATYGTTGACATYCAAGC</td>
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<tr>
<td></td>
<td>Tubu-1538R</td>
<td>TAYTGYGTGATYTCGCTRACY</td>
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<tr>
<td></td>
<td>Tubu-63F</td>
<td>CAAATWGGYGCGAARTYTTGGA</td>
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<td></td>
<td>Tubu-3R</td>
<td>TCGTCCATACCTTTACCGTACCAGT</td>
</tr>
<tr>
<td>CCT7</td>
<td>TBCCT35F</td>
<td>TGAAGGARGGNACNGAAYACWTTCYACG</td>
</tr>
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<td></td>
<td>TBCCTR0</td>
<td>GTYTCRTCDATDWSNAGNACHWGGCAGNGCNGCYTCDGTNGC</td>
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<td>TBCCT70F</td>
<td>CAAATYATYAYAAAYATWAAAGCCTGYYC</td>
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<td></td>
<td>TBCCT1519R-3</td>
<td>KTYYTYTNNACMANNHBHDGTYCCADATRCA</td>
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</table>
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Authors’ addresses: Haruyuki Hirata, Mari Maeda, Michio Jinnai, Kohei Fujisawa, and Chiaki Ishihara, School of Veterinary Medicine, Rakuno Gakuen University, Ebetsu, Japan, E-mails: hirata@rakuno.ac.jp, s20603057@stu.rakuno.ac.jp, s20741005@stu.rakuno.ac.jp, s20741003@stu.rakuno.ac.jp, and ishihara@rakuno.ac.jp. Satoru Kawai, Laboratory of Tropical Medicine and Parasitology, Dokkyo Medical University, Tochigi, Japan, E-mail: skawai@dokkyomed.ac.jp. Yuko Katakai, Corporation for Production and Research of Laboratory Primates, Hokkaido University, Sapporo, Japan, E-mail: kikai@hokkaido-primate.or.jp. Kenji Hikosaka and Kazuyuki Tanabe, Laboratory of Biomedical Innovation, Tsukuba, Ibaraki, Japan, E-mail: yasutomi@nibio.go.jp.

REFERENCES


