

Bartonella quintana, Lice, and Molecular ToolsPHILIPPE PAROLA, PIERRE-EDOUARD FOURNIER, AND DIDIER RAOULT¹

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We read with interest but question the recent article by Sasaki et al. (2006) that reported the molecular detection by nested polymerase chain reaction (PCR) of the agent of trench fever, *Bartonella quintana*, in human body louse, *Pediculus humanus humanus* L., the known vector, as well as in human head louse, *Pediculus humanus capitis* De Geer, all collected on Nepalese children. Head lice are widely distributed among human populations, whereas body lice parasitism is linked to poverty, cold climate, and lack of hygiene. Therefore, all implications of head lice as potential vectors of pathogens need to be carefully controlled by rigorous methods. Several imprecisions and inconsistencies limit the value of this study. First, no information is given about the controls, and particularly the negative controls that were used, although these are essential in interpreting positive PCR results. Nested PCR is hampered by a high risk of contamination (Millar et al. 2002). Alternatively, nested PCR techniques using a closed assay or single-use primers without positive controls reduce such a risk (Raoult et al. 2006). In this study, negative controls are neither described in the Materials and Methods nor shown on any gel in the Results. Second, the identification of lice in this article was based on the comparison of the length of the same parts of the legs of recently blood-fed adults. The validity of the morphological differentiation between head and body lice, based on these subtle differences, is dubious. Furthermore, the authors write that they amplified 18S rRNA gene fragments to control the extraction efficacy and the presence of PCR inhibitors. It is well known that patients who are heavily infested with body lice also may carry these ectoparasites on the head, and we often noticed this fact when examining homeless people in Marseille (Fournier et al. 2002; Brouqui et al. 2005). Therefore, we believe the results presented by Sasaki et al. (2006) may be due to contamination or confusion in the identification of the lice. To avoid mistakes, it is best to study lice on people

who are infested with only one type of louse: either head or body (Yong et al. 2003). In our laboratory, *B. quintana* has never been detected in thousands of head lice, collected throughout the world. Finally, it should be remembered that the molecular detection of bacteria in any arthropod does not necessarily indicate that the arthropod is a potential vector or plays a role in the transmission of the bacteria. Despite the authors' conclusion that their results "demonstrate" that head lice may play a role in the transmission of trench fever, we believe that the demonstration is not sufficiently rigorous or at least not rigorously presented. These data need further confirmation.

References Cited

- Brouqui, P., A. Stein, H. Tissot-Dupont, P. Gallian, S. Badiaga, J.-M. Rolain, J.-L. Mege, B. La Scola, P. Berbis, and D. Raoult. 2005. Ectoparasitism and vector-borne diseases in 930 homeless people from Marseilles. *Medicine (Balt.)* 84: 61–68.
- Fournier, P.-E., J.-B. Ndhokubwayo, J. Guidran, P. J. Kelly, and D. Raoult. 2002. Human pathogens in body and head lice. *Emerg. Infect. Dis.* 8: 1515–1518.
- Millar, B. C., J. Xu, and J. E. Moore. 2002. Risk assessment models and contamination management: implications for broad-range ribosomal DNA PCR as a diagnostic tool in medical bacteriology. *J. Clin. Microbiol.* 40: 1575–1580.
- Raoult, D., O. Dutour, L. Houhamdi, R. Jankauskas, P.-E. Fournier, Y. Ardagna, M. Drancourt, M. Signoli, V. D. La, Y. Macia, and G. Aboudharam. 2006. Evidence for louse-transmitted diseases in soldiers of Napoleon's Grand Army in Vilnius. *J. Infect. Dis.* 193: 112–120.
- Sasaki, T., S. K. Poudel, H. Isawa, T. Hayashi, N. Seki, T. Tomita, T., K. Sawabe, and M. Kobayashi. 2006. First molecular evidence of *Bartonella quintana* in *Pediculus humanus capitis* (Phthiraptera: Pediculidae), collected from Nepalese children. *J. Med. Entomol.* 43: 110–112.
- Yong, Z., P.-E. Fournier, E. Rydkina, and D. Raoult. 2003. The geographical segregation of human lice preceded that of *Pediculus humanus capitis* and *Pediculus humanus humanus*. *C. R. Biol.* 326: 565–574.

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