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Evolution of nitrogen-fixing endosymbionts of termite-gut protists

Yuichi Hongoh

Wood-feeding termites generally need taking up nitrogen from the atmosphere with the aid of N2-fixing gut bacteria to balance the low nitrogen content in their food materials (1). One of such N2-fixing bacteria, *Candidatus Azobacteroides pseudotrichonymphae* (order Bacteroidales), is an intracellular symbiont of the cellulolytic protist *Pseudotrichonympha grassii* in the gut of the Formosan subterranean termite *Coptotermes formosanus*. *A. pseudotrichonymphae* predominates in the gut microbiota, accounting for more than 50% of the total bacterial cells (2), and the *Azobacteroides* bacteria, the *Pseudotrichonympha* protists, and the host rhinotermitid termites have strictly cospeciated (3). Although it is difficult to examine the functions of *Azobacteroides* because of their unculturability, our genome analysis of *A. pseudotrichonymphae* has previously suggested that the bacterium fixes nitrogen and synthesizes various amino acids and cofactors. In addition, we suggested that the bacterium potentially utilizes the waste products of the host protists: it recycles urea and ammonia as nitrogen sources and consumes hydrogen as an energy source (4, 5). Thus, it is likely that the bacterium and the hosts have a close mutualistic relationship. In my talk, I will present our recent results of comparative genomics of *Azobacteroides* endosymbionts of the *Pseudotrichonympha* protists in the gut of several rhinotermitid termite species. We performed whole genome amplification and successfully reconstructed complete or nearly complete genomes of these unculturable bacteria. Our study demonstrates how the ecology and evolution of the host termites are in harmony with the functions of gut bacteria.

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