

Abstract

Listeria monocytogenes is a human food borne pathogen that may lead, in particular in immunocompromised individuals, to a severe disease characterized by septicemias, meningitis, meningo-encephalitis and abortions. The study of the cell biology of *Listeria* infectious process provided insights in the way bacteria manipulate the host and revealed unsuspected functions of cellular proteins. To cause infection pathogens interfere with crucial host intracellular pathways, different pathogens often hijacking the same signaling pathways. In particular, host phosphorylation cascades are preferential targets of infecting bacteria.

In this study, using *L. monocytogenes* as a pathogen model, we showed that eukaryotic cells present a variable protein phosphorylation pattern upon infection. We addressed in particular the tyrosine-phosphorylated protein profile triggered by *Listeria* infection and identified two cytoskeletal proteins, Myosin 9 (Myh9) and Cytokeratin 18 (CK18), differentially tyrosine-phosphorylated in response to *Listeria* uptake. We demonstrated that Myh9 was not only tyrosine-phosphorylated over the time of infection, but was also recruited with actin at the bacteria entry site. In addition, we were able to show that the inhibition of Myh9 activity blocked *Listeria* entry into non-phagocytic cells. Surprisingly, the reduction of Myh9 expression using RNAi techniques resulted in an increased *Listeria* uptake.

Together these results point to the role of a novel myosin class in the internalization of *Listeria*, correlating for the first time myosin post-translational modifications and *Listeria* infection. We also show here that CK18 is enriched in the vicinity of entering *Listeria* into epithelial Caco-2 cells. Moreover, our preliminary results indicate a reduced entry of *Listeria* in cells showing low levels of CK18 expression. The identification of Myh9 and CK18 tyrosine-phosphorylated residues during infection and their direct mutagenesis would help to unravel the direct role of these specific tyrosine phosphorylations on bacteria internalization. This work identified two novel host proteins involved in the *Listeria* infectious process and should contribute to the identification of new signaling cascades hijacked by pathogens to promote their own internalization.