Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Microbiology

Actin-based motility and cell-to-cell spread of bacterial pathogens

Rebecca L Lamason and Matthew D Welch

Subversion of the host actin cytoskeleton is a critical virulence mechanism used by a variety of intracellular bacterial pathogens during their infectious life cycles. These pathogens manipulate host actin to promote actin-based motility and coordinate motility with cell-to-cell spread. Growing evidence suggests that the tactics employed by pathogens are surprisingly diverse. Here, we review recent advances suggesting that bacterial surface proteins exhibit divergent biochemical mechanisms of actin polymerization and recruit distinct host protein networks to drive motility, and that bacteria deploy secreted effector proteins that alter host cell mechanotransduction pathways to enable spread. Further investigation into the divergent strategies used by bacterial pathogens to mobilize actin will reveal new insights into pathogenesis and cytoskeleton regulation.

Address
Department of Molecular and Cell Biology, University of California, Berkeley, CA 94720, USA

Corresponding authors: Lamason, Rebecca L (rlamason@berkeley.edu) and Welch, Matthew D (welch@berkeley.edu)

Current Opinion in Microbiology 2017, **35**:48–57

This review comes from a themed issue on **Host–microbe interactions: bacteria**

Edited by **Samuel Miller** and **Renée Tsolis**

http://dx.doi.org/10.1016/j.mib.2016.11.007

Introduction

In eukaryotic cells, the actin cytoskeleton regulates a variety of functions. For example, it provides structural integrity at cell–cell junctions to maintain tissue integrity, and dynamically reorganizes to promote the formation of membrane extensions or invaginations during cell migration and intracellular trafficking [1–3]. Due to its importance in these diverse cellular processes, the actin cytoskeleton is also a critical target of intracellular bacterial pathogens. Many pathogens hijack actin at different steps of their life cycle, and investigating these processes has revealed new ways in which host cells regulate actin cytoskeleton dynamics in uninfected settings [4].

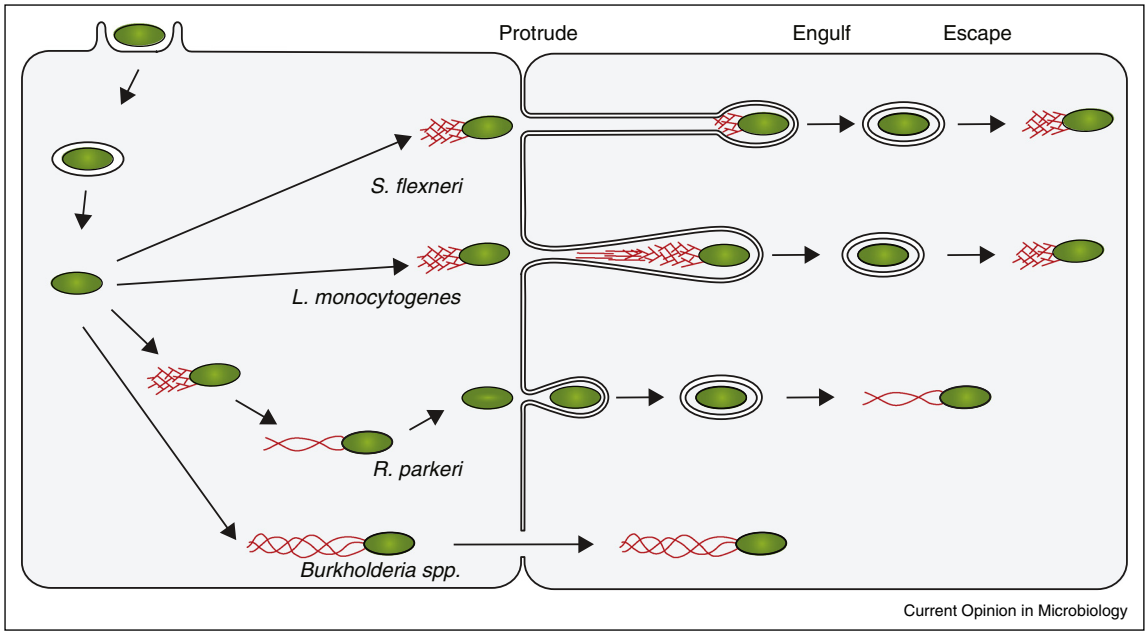
In this review, we will discuss recent advances in our understanding of the molecular mechanisms by which intracellular bacterial pathogens exploit actin. We will focus on pathogens within four genera, including *Listeria monocytogenes*, *Shigella flexneri*, *Burkholderia* spp. in the pseudomallei group, and spotted fever group (SFG) *Rickettsia* spp. These bacteria are evolutionarily diverse — *Listeria* spp. are Gram-positive firmicutes, whereas the others are Gram-negative alphaproteobacteria (*Rickettsia* spp.), betaproteobacteria (*Burkholderia* spp.) or gammaproteobacteria (*Shigella* spp.). They are also transmitted by different routes, and cause a spectrum of diseases including listeriosis (*L. monocytogenes*), shigellosis (*S. flexneri*), meliodosis (*B. pseudomallei*), glanders in equines (*B. mallei*), and spotted fever and eschar-associated rickettsioses (*Rickettsia* spp.) [5]. Despite their overall diversity, these pathogens share a common mechanism of infection. In particular, they invade non-phagocytic cells and escape the phagosome into the cytosol where they polymerize actin filaments to generate actin 'comet' tails on their surface to drive movement. Actin-based motility propels the bacteria through the cytosol and enables spread into neighboring cells (Figure 1) [6–8].

We will focus on two themes that have emerged recently. The first is that, despite common features of infection, recent work has revealed surprising differences in the molecular mechanisms of actin-based motility. Older work showed a critical role for the host Arp2/3 complex and its nucleation promoting factors (NPFs) in actin assembly [9,10], but we are now learning that diverse biochemical mechanisms of actin polymerization are used by pathogens, resulting in divergent actin filament organization and parameters of motility. We are also learning that various host proteins regulate bacterial motility. The second emerging theme is that the parameters and mechanisms of spread are also quite diverse between pathogens, with differential dependence on actin-based motility and distinct ways of remodeling the actin cytoskeletal network at cell–cell junctions. Though more work is needed to fully elucidate the molecular mechanisms and key players involved in motility and spread, we are beginning to understand that these are dynamic and complicated processes coordinated by a network of host and bacterial factors.

Diverse biochemical mechanisms of actin-based motility

Once inside host cells, the pathogens highlighted in this review polymerize actin on their surface to rocket through

Figure 1



Life cycles of intracellular bacterial pathogens that harness actin-based motility to enable cell-to-cell spread. The cartoon depicts the intracellular life cycles of the pathogens discussed in this review. After invading bacteria are phagocytosed and escape the phagosome, they enter the host cell cytosol, where they polymerize actin using distinct mechanisms and undergo actin-based motility, forming actin comet tails with different filament organizations. *R. parkeri*, a representative of the SFG of *Rickettsia* spp., undergo two temporally segregated and biochemically distinct phases of actin-based motility, as depicted. All of these pathogens also undergo diverse pathways of cell-to-cell spread via protrusion- and vesicle-mediated transfer (for *S. flexneri*, *L. monocytogenes*, and *Rickettsia* spp.), or direct cell–cell fusion (for *Burkholderia* spp). Actin, red; bacteria, green.

the cytoplasm, leaving in their wake actin comet tails. Early work showed that several bacterial species hijack the host Arp2/3 complex to polymerize actin tails consisting of branched filament networks, leading to motility characterized by curved or meandering paths (Figure 2) [9,11•]. At the molecular level, the bacterial surface proteins ActA from *L. monocytogenes*, BimA from *B. thailandensis* (BtBimA) and RickA from SFG rickettsiae mimic host nucleation promoting factors (NPFs) to activate the Arp2/3 complex [12–17]. In contrast, *S. flexneri* IcsA (also called VirG) recruits the host NPF N-WASP to the bacterial pole to activate Arp2/3 [18,19]. These early studies supported the idea that the Arp2/3 complex was crucial for pathogen motility, and many assumed this mechanism was conserved across all species.

It is now becoming clear, however, that hijacking the Arp2/3 complex is not the only pathway bacteria use to polymerize actin. Recent work has uncovered that actin assembly by different pathogens occurs by vastly different biochemical mechanisms, resulting in differences in the types of actin networks generated and the parameters of motility exhibited (Figure 2). For example, in addition to RickA, SFG *rickettsiae* also express Sca2, a mimic of host formin proteins, which directly nucleates actin and promotes filament elongation by processively associating with the barbed end of an actin filament and preventing host capping protein activity [20,21•]. In contrast, BimA proteins from *B. pseudomallei* (BpBimA) and *B. mallei* (BmBimA) mimic host Ena/VASP actin polymerases in their ability to promote filament elongation and bundling [22•]. Actin tails formed by Sca2- and BpBimA/BmBimA-mediated actin assembly are composed of long bundled actin filaments, leading to straighter paths of motility (Figure 2a) [11•,22•]. The existence of bacterial formin and Ena/VASP mimics underscores the surprising variety of actin-based motility mechanisms bacteria have evolved by coopting diverse host actin assembly strategies.

It remains unclear, though, how the evolution of distinct actin polymerization mechanisms influences virulence. Interestingly, BimA from the non-pathogenic *Burkholderia* species *B. thailandensis* uses an Arp2/3-dependent polymerization pathway, whereas BimA from the pathogenic species *B. pseudomallei* and *B. mallei* use an Ena/VASP mimic (Figure 2b), suggesting these differences could contribute to virulence. Moreover, studies of Australian *B. pseudomallei* strains that express either BpBimA or BmBimA revealed that there is an association between BmBimA expression and neurological melioidosis, and BpBimA and pneumonia [23••], again suggesting a correlation between differences in actin assembly and virulence. It remains to be determined whether and how the