Abstract

Bordetella pertussis is a strictly human pathogen and causative agent of infectious respiratory disease called whooping cough. In order to establish successful infection and colonization of the host, *B. pertussis* uses a broad spectrum of virulence factors such as adhesins (filamentous hemagglutinin, pertactin, and fimbriae) and toxins (adenylate cyclase and pertussis toxins). In addition, the type 3 secretion system (T3SS) was also found in the genus *Bordetella*. In connection to our previous characterisation of *B. pertussis* strain lacking the gene encoding RNA chaperone Hfq (Δ*hfq*), which proved that Hfq is required for T3SS functionality, the recombinant T3SS proteins BopB, BopD, BopC and BopN were purified to homogeneity. Next, the specific antibodies were obtained using purified recombinant proteins in order to study the production of the T3SS components in *B. pertussis*. Using refined anti-BopC antibodies it was for the first time shown that laboratory-adapted *B. pertussis* strain secretes BopC protein into medium. The recombinant translocators BopB and BopD were also used to examine their pore-forming activity using planar black lipid membranes. Based on the characterisation of *hfq* deletion mutant, having impaired production of membrane proteins when compared to the wild type, mass spectrometry analysis was used to identify proteins with different abundance. The outcome of this analysis was in agreement with our previous transcriptomic data.

Key words: type 3 secretion system (T3SS), BopB, BopC, BopD, BopN, pore-forming activity, membrane isolation, Hfq, infection