The role of ActA in peptidoglycan remodelling in *Listeria monocytogenes*

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Listeria monocytogenes is a food-borne bacterial pathogen, the causative agent of human listeriosis. It may cause abortion in pregnant women, septicaemia, endocarditis and meningitis in elderly people and immunocompromised patients. *L. monocytogenes* has many virulence factors that enable its replication in macrophages and the escape from the phagolysosome to the cytoplasm. One of these virulence factors is the actin-assembly inducing protein, ActA, that is essential for *L. monocytogenes* intra- and intercellular motility. Recently, the ActA protein has been shown to regulate peptidoglycan (PG) biosynthesis during *L. monocytogenes* replication in macrophages. However, the exact mechanism for this phenomenon is unknown. The central hypothesis of the present study is that ActA possesses peptidoglycan hydrolysing activity. To address this hypothesis three different His-tagged forms of ActA have been expressed in *Escherichia coli*. All versions have been successfully purified using affinity chromatography, gel-filtration and their identity has been confirmed by mass-spectrometry. Peptidoglycan hydrolysing activity of these proteins has been assessed by application of zymography and digestion of FITC-labelled peptidoglycan. One domain showed significant peptidoglycan-hydrolysing activity as judged by zymography and digestion of FITC-labelled peptidoglycan. Candidate catalytic residues are currently being identified by application of bioinformatics and their function will be verified by site directed mutagenesis. Future experiments such as analysis of muropeptides released from PG by ActA, complementation studies and pull-down assays will shed light on the function of this protein in peptidoglycan remodelling.

Speaker Biography

Ohoud S Alhumaidan is a 3rd year PhD student in infection, immunity and inflammation department at University of Leicester. She is working under the supervision of Dr. Galina Mukamolova and professor Peter Andrew. Her doctoral research investigates the role of specific protein in *listeria monocytogenes*. She is a member of the microbiology society and she holds master's degree in microbiology from King Saud University, Saudi Arabia, in isolation and characterisation of nasal carriage MRSA among health care staff in a teaching hospital.

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