

Mini-Review

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Lipoprotein synthesis in mycobacteria

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Lipoproteins are a functionally diverse class of secreted bacterial proteins characterized by an N-terminal lipid moiety. The lipid moiety serves to anchor these proteins to the cell surface. Lipoproteins are synthesized as pre-prolipoproteins and mature by post-translational modifications. The post-translational modifications are directed by the lipobox motif located within the signal peptide. Enzymes involved in lipoprotein synthesis are essential in Gram-negative bacteria but not in Gram-positive bacteria. Inactivation of genes involved in lipoprotein synthesis attenuates a variety of Gram-positive pathogens, including *Mycobacterium tuberculosis*. The attenuated phenotype of these mutants indicates an important role of lipoproteins and lipoprotein synthesis in bacterial virulence. *M. tuberculosis*, the causative agent of tuberculosis, is one of the most devastating pathogens in the world. This article reviews recent findings on the synthesis, localization and function of lipoproteins in mycobacteria.

Introduction

Lipoproteins are a functionally heterogeneous class of proteins universally present in bacteria; typically between 1 % and 3 % of bacterial genomes encode lipoproteins (Babu *et al.*, 2006; <http://www.mrc-lmb.cam.ac.uk/genomes/dolop/>). Lipoproteins represent a subgroup of secreted proteins characterized by the presence of a lipobox. The lipobox motif is located in the C-terminal part of the leader peptide and consists of four amino acids [LVI/ASTVI/GAS/C] (Babu *et al.*, 2006). This motif functions as a recognition signal for lipid modification, which is made on the conserved and essential cysteine residue. Precursor lipoproteins are mainly translocated in a Sec-dependent manner across the plasma membrane and are modified subsequently. Recent investigations indicate that lipoproteins may also be translocated by the twin-arginine translocation (Tat) system (McDonough *et al.*, 2005). However, this pathway presumably is of minor importance because the number of lipoproteins translocated via the Tat transporter is rather small. Modification of precursor proteins is mediated by the consecutive activity of three enzymes: phosphatidylglycerol–pre-prolipoprotein diacylglyceryl transferase (Lgt), prolipoprotein signal peptidase/signal peptidase II (LspA) and phospholipid–apolipoprotein *N*-acyltransferase (Lnt) (Fig. 1). While Lgt and LspA are universally present in bacteria, Lnt has been reported to be restricted to Gram-negatives (Wu, 1996). The lipid residue covalently linked to the conserved cysteine moiety is thought to allow for anchoring of proteins in biological membranes by means of hydrophobic interaction. In Gram-positive bacteria, cell-associated lipoproteins are found in the plasma membrane. In Gram-negative bacteria the

majority of cell-associated lipoproteins are found in the outer membrane – only about 10 % stay anchored in the plasma membrane. Lipoproteins may be divided into five general groups according to their function in adhesion and invasion, cell wall synthesis, nutrient uptake, degradative processes, and sensing and transmembrane signalling (Sutcliffe & Russell, 1995). Enzymes involved in lipoprotein synthesis (Lgt, LspA, Lnt) are essential in Gram-negative bacteria but not in Gram-positive bacteria (Wu, 1996; Leskela *et al.*, 1999).

Mycobacteria belong to the group of GC-rich, Gram-positive bacteria, although the cell envelope of these bacteria is rather complex and in some respects resembles the cell envelope of Gram-negatives (Brennan & Nikaido, 1995). The lipid-rich outer layer in mycobacteria formed by mycolic acids and phospholipids is analogous to the outer membrane of Gram-negative bacteria. The genus *Mycobacterium* consists of more than 120 species (Rogall *et al.*, 1990). The genome sequences of ten mycobacterial species have been determined or sequencing is near to completion (<http://www.sanger.ac.uk>; <http://www.tigr.org>). With respect to pathogenicity mycobacteria may be divided into obligate pathogens, opportunistic pathogens or non-pathogenic species. *Mycobacterium tuberculosis* and *Mycobacterium leprae*, the causative agents of tuberculosis and leprosy, are major human pathogens. Opportunistic infections, in particular in patients with immune suppression, may be due to a variety of mycobacteria, e.g. *Mycobacterium avium*–*Mycobacterium intracellulare*, *Mycobacterium marinum*, *Mycobacterium kansasii* and *Mycobacterium chelonae*. *Mycobacterium smegmatis*, a non-pathogenic, fast-growing mycobacterium, has been

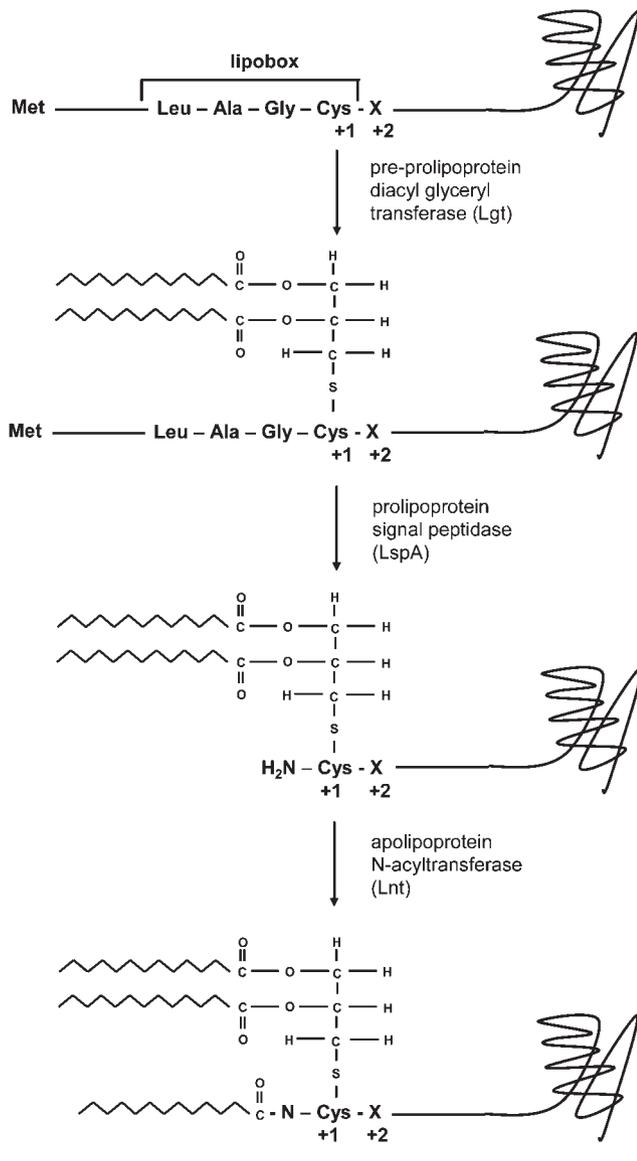


Fig. 1. Biosynthesis of bacterial lipoproteins. Precursor lipoproteins are post-translationally modified by phosphatidylglycerol-pre-prolipoprotein diacylglyceryl transferase (Lgt) and prolipoprotein signal peptidase (LspA). In Gram-negative bacteria and probably also in mycobacteria, lipoproteins are further processed by apolipoprotein *N*-acyltransferase (Lnt).

established as a workhorse in mycobacterial genetics and biochemistry. Bioinformatic analyses suggest the presence of at least 48 lipoproteins but potentially twice as many in the genome sequence of *M. tuberculosis* (Sutcliffe & Harrington, 2004); similar numbers are found in other mycobacteria (Table 1). Experimental data concerning mycobacterial lipoprotein function are rare. Nearly half of the annotated *M. tuberculosis* lipoproteins do not share conserved domains with proteins outside the genus *Mycobacterium* and thus represent unique proteins of mycobacteria (Sutcliffe & Harrington, 2004). We here review recent findings on the synthesis, localization and function of lipoproteins in

mycobacteria; emphasis is given to *M. tuberculosis* and *M. smegmatis*.

Mycobacterial lipoprotein synthesis

Lgt. Lipoproteins are synthesized as pre-prolipoproteins and mature by post-translational modifications. Lipoprotein-modifying enzymes are membrane-integral proteins located in the plasma membrane. The first step of lipoprotein synthesis is conferred by Lgt, which adds a diacylglycerol residue to the thiol group of the universally conserved cysteine within the lipobox (Fig. 1). In *Escherichia coli*, the lipid moiety of lipoproteins is derived from the membrane lipid phosphatidylglycerol (Wu, 1996). Phosphatidylglycerol is not a homogeneous entity, as its acyl chains may be composed of 16–20 carbon atoms. In addition, the acyl chains may be saturated or unsaturated. So far, lipid moieties of mycobacterial lipoproteins have not been determined at the molecular level. However, in analogy to *E. coli* it may be hypothesized that the lipid moieties reflect the composition of mycobacterial membrane phospholipids. Therefore, despite identical protein moieties lipoproteins may vary to some extent.

Lipid modification of proteins can be demonstrated by labelling with radioactive palmitate and by detergent solubility. Early investigations exploited incorporation of [¹⁴C]palmitic acid to demonstrate lipoprotein synthesis in *M. tuberculosis* (Young & Garbe, 1991). Radioactive labelling was also used to demonstrate protein lipidation in *M. smegmatis* (Kriakov *et al.*, 2003). *M. tuberculosis* Lgt (Rv1614) is composed of 468 aa (mol. mass 50.4 kDa) and thus considerably larger than its *E. coli* homologue (291 aa). Compared to *E. coli* the *M. tuberculosis* Lgt, as well as Lgt of other members of the order *Actinomycetales*, is characterized by an additional C-terminal domain of unknown function. The isoelectric points of *M. tuberculosis* Lgt (pI 4.45) and *E. coli* (pI 9.66) differ greatly. The high pI of the *E. coli* enzyme is assumed to be important for interaction with acidic phospholipids by ionic as well as hydrophobic interactions (Wu, 1996). The low pI of the *M. tuberculosis* Lgt enzyme is due to the unusually high aspartate and glutamate content of the C-terminal extension. In the *M. smegmatis* genome MSMEG3232 is annotated as Lgt. MSMEG3232 is composed of 612 aa (mol. mass 64.6 kDa) and like its *M. tuberculosis* homologue has a low pI (3.92). The functional consequences of the dramatic pI differences between the *E. coli* and the mycobacterial Lgt remain to be elucidated. In *M. smegmatis* a second ORF, MSMEG5388, is annotated as a putative Lgt. This ORF encodes a protein of 261 aa (mol. mass 31.0 kDa) with a pI of 9.38. However, a multiple sequence alignment revealed that the protein encoded by MSMEG5388 differs from the Lgt consensus sequence at several conserved residues (data not shown).

LspA. Lgt-modified prolipoproteins are further processed by LspA. LspA cleaves off the signal peptide directly in front of the modified cysteine. LspA knock-out mutants therefore accumulate prolipoproteins (Fig. 1).

Table 1. Lipoproteins in representative mycobacterial genomes

LipoP was used as a tool to predict lipoprotein signal peptides (<http://www.cbs.dtu.dk/services/LipoP/>; Juncker *et al.*, 2003). Protein sequences were downloaded from the TIGR comprehensive microbial resource database (<http://www.tigr.org>), except for *M. marinum*, which was downloaded from the server of the Sanger Institute (<http://www.sanger.ac.uk>). The lower number of predicted lipoproteins in our search as compared to previous reports (e.g. Sutcliffe & Harrington, 2004) is due to the more restrictive LipoP algorithm.

Species	No. of predicted lipoproteins	No. of ORFs	Relative no. of lipoproteins (%)
<i>M. tuberculosis</i>	48	3918	1.2
<i>M. leprae</i>	25	1604	1.6
<i>M. avium</i>	75	4350	1.7
<i>M. marinum</i>	97	5485	1.8
<i>M. smegmatis</i>	61	6776	0.9

Due to the presence of the leader peptide, prolipoproteins have a slightly (2–3 kDa) higher molecular mass than the mature lipoproteins. Accumulation of prolipoproteins was demonstrated in *M. tuberculosis* *lspA* (Rv1539) (Sander *et al.*, 2004; Banaiee *et al.*, 2006) and recently in *M. smegmatis* *lspA* (MSMEG3181) knock-out mutants (M. Rezwani, A. Tschumi, T. Grau, S. Kuhn, P. Keller, B. Springer, E. C. Böttger & P. Sander, unpublished data). Inactivation of *M. tuberculosis* LspA by allelic replacement revealed an essential role of lipoprotein synthesis in the pathogenesis of *M. tuberculosis*. An *M. tuberculosis* *lspA* knock-out mutant exhibited reduced multiplication in the mouse macrophage cell line J774, complete absence of lung pathology and a 3–4 log reduced number of c.f.u. in a mouse model of tuberculosis infection (Sander *et al.*, 2004). However, the molecular mechanisms underlying *M. tuberculosis* *lspA* attenuation remain to be determined.

Lipoproteins are potent agonists of TOLL-like receptor (TLR) 2, and extracts of an *M. tuberculosis* LspA knock-out mutant failed to induce a TLR2 response in the TLR2-reporter cell line HEK293. However, TLR-dependent activation in macrophages by entire mycobacteria or whole-cell lysates was not affected by LspA inactivation (Banaiee *et al.*, 2006), indicating the existence of TLR ligands other than mature lipoproteins. The redundancy of TLR agonists makes it unlikely that attenuation of the *lspA* knock-out mutant is due to alterations in TLR signalling.

Globomycin, a cyclic peptide produced by several *Streptomyces* species, is a potent and specific inhibitor of lipoprotein signal peptidases in different bacterial species. Due to the essentiality of lipoprotein-synthesizing enzymes, globomycin exerts a bactericidal effect in *E. coli* (Inukai *et al.*, 1978). Inhibition of lipoprotein maturation by globomycin, as indicated by accumulation of precursor lipoproteins, has also been shown for Gram-positive bacteria (Harrington *et al.*, 2000). Due to the severe attenuation of the *M. tuberculosis* *lspA* knock-out mutant, LspA has been proposed as a putative mycobacterial drug target and exploitation of globomycin as a prototype inhibitor for

further drug development has been suggested (Sander *et al.*, 2004). Promising antibacterial activity of globomycin derivatives has recently been demonstrated in Gram-positive bacteria (Kiho *et al.*, 2004), but their activity towards mycobacteria remains to be determined.

Lnt. In Gram-negative proteobacteria, but not in Gram-positive bacteria, LspA-processed lipoproteins are further modified by Lnt (Wu, 1996). Lnt adds a third acyl residue to the amino group of the modified cysteine. *In vitro* and *in vivo* studies have indicated that any of the three major phospholipids in the *E. coli* cell envelope (phosphatidylethanolamine, phosphatidylglycerol, cardiolipin) can serve as an acyl donor (Wu, 1996). In *E. coli*, Lnt modification is a prerequisite for transport of lipoproteins across the periplasm. Based on the assumption that N-acylation of lipoproteins is required for transport into the mycolic acid layer, the presence of Lnt homologues in mycobacteria may be postulated. Multiple sequence alignments (data not shown) confirmed early *in silico* findings (Gurcha *et al.*, 2002; Baulard *et al.*, 2003) that Lnt homologues are present in mycobacteria. *M. tuberculosis* Rv2051c encodes a two-domain protein, where the N-terminal part shows similarity to *E. coli* Lnt. The C-terminal part of the protein encodes a polyprenylmonophosphomannose (Ppm) synthase, which transfers mannose from GDP-mannose to endogenous polyprenyl phosphates, an important metabolic intermediate in the synthesis of the mycobacterial cell wall constituents lipomannan and lipoarabinomannan (LAM). The Lnt-homology domain of Rv2051c enhances the Ppm synthase activity of the C-terminal domain, while apo-lipoprotein N-acyltransferase activity remains to be demonstrated (Baulard *et al.*, 2003). Besides the N-terminal domain of Rv2051c, ORF Rv2262 has lower but still significant homology to *E. coli* Lnt. Again, an enzymic function remains to be determined. In *M. smegmatis*, orthologues of the two domains of *M. tuberculosis* Rv2051c are encoded by two distinct ORFs, Msppm1 and Msppm2, of which Msppm2 (MSMEG3863) corresponds to Lnt. Thus, the *M. tuberculosis* Lnt ORF encodes a significantly

larger protein (874 aa) compared to *M. smegmatis* (654 aa) and *E. coli* (512 aa). Lnt homologues have been found not only in mycobacteria but also in other actinobacteria, including *Streptomyces* and *Corynebacterium*. A comprehensive analysis of fully sequenced genomes of bacteria representing different phyla suggests that Lnt homologues are present in micro-organisms with a complex cell envelope (unpublished observations) but not in ordinary Gram-positive bacteria.

The presence of Lnt homologues and the localization of lipoproteins in the mycobacterial cell wall (see below) suggests that a transport system for lipoproteins into the mycolic acid layer should exist – homologous or analogous to the *E. coli* Lol system. The *E. coli* Lol system consists of (i) an ABC-transporter complex of three proteins (LolC, LolD and LolE), which releases lipoproteins from the plasma membrane; (ii) a periplasmic chaperone (LolA); and (iii) a receptor in the outer membrane (LolB), which is itself a lipoprotein. *E. coli* lipoproteins are translocated to the outer membrane unless they possess a Lol-avoidance signal, i.e. an aspartate at position +2 (Narita *et al.*, 2004). More complex lipoprotein transport signals may be present in other bacterial species (Schulze & Zuckert, 2006). Protein sequence alignments so far have not identified Lol homologues in *M. tuberculosis*. However, recent structural analysis of *M. tuberculosis* lipoprotein LppX revealed structural homologies to *E. coli* LolA and LolB, although the primary substrate seems to be phthiocerol dimycoserate rather than lipoproteins (Sulzenbacher *et al.*, 2006).

Lipoprotein localization

The function of a protein depends on its correct localization and vice versa. In Gram-positive bacteria, cell-associated lipoproteins stay anchored in the plasma membrane. In Gram-negative bacteria, the vast majority of cell-associated lipoproteins are released from the plasma membrane and are anchored to the outer membrane (Tokuda & Matsuyama, 2004). Despite recent progress in subcellular fractionation of mycobacteria (Mawuenyega *et al.*, 2005; Rezwani *et al.*, 2006), localization of mycobacterial lipoproteins has rarely been addressed. *M. tuberculosis* LprG and *M. smegmatis* PhoA were shown to be anchored in the cell envelope (Kriakov *et al.*, 2003); likewise *M. tuberculosis* Mpt83 was shown to be cell surface associated by electron microscopy and fluorescence cytometry (Vosloo *et al.*, 1997; Harboe *et al.*, 1998). Proteomic analyses of fractionated *M. tuberculosis* extracts identified and located 28 putative lipoproteins (Mawuenyega *et al.*, 2005). Six lipoproteins were found in the cell wall fraction, ten in the plasma membrane fraction and seven in the cytosolic fraction; five lipoproteins were observed in two or in all three fractions. Localization of mature lipoproteins in the cytosol is questionable and these lipoproteins may represent recently synthesized (pre-pro-) proteins not yet secreted. Subcellular fractionation of *lgt*, *lspA* and *lnt* mutants expressing recombinant proteins will help to elucidate the mechanisms underlying mycobacterial lipoprotein sorting.

Lipoproteins

In an excellent review Sutcliffe & Harrington (2004) discussed the function of individual *M. tuberculosis* lipoproteins in great detail. We therefore limit our discussion to recent findings. Most mycobacterial lipoproteins have been predicted by a bioinformatic approach, i.e. identification of the lipobox consensus sequence within a typical signal peptide (Sutcliffe & Harrington, 2004). Experimentally investigated lipoproteins often are immunodominant antigens. However, pleiotropic and seemingly antagonistic, i.e. pro- and anti-inflammatory effects, make it difficult to discern the mechanisms by which these proteins contribute to immunopathogenesis (Karakousis *et al.*, 2004). Investigations with *M. leprae* LpK variants differing in the length of the peptide and the presence or absence of the lipid moiety, revealed that both acyl residues and peptide sequences are required for elicitation of an immune response. The inability of a non-acylated 27 kDa antigen of *M. tuberculosis* to induce IFN- γ secretion corroborates the importance of the lipid moiety for inducing an immune response (Hovav *et al.*, 2004).

Targeted gene inactivation of individual lipoprotein genes, transposon site hybridization mutagenesis and vaccination studies corroborate early findings on the overall importance of lipoproteins in the immunopathogenesis of infection with *M. tuberculosis*. Several putative lipoproteins, e.g. LppP (Rv2330c), LprO (Rv2290), LprK (Rv0173), LpqT (Rv1016c), LpqY (Rv1235), LpqZ (Rv1244), LprG (Rv1411) and LppX (Rv2945c), are required for optimal growth *in vivo* (Rengarajan *et al.*, 2005; Sasseti & Rubin, 2003; Bigi *et al.*, 2004). Some of these *M. tuberculosis* lipoproteins have homologues in mycobacteria, others represent signature proteins for the *Corynebacterium*, *Mycobacterium*, *Nocardia* (CMN) subgroup of the actinobacteria (Gao *et al.*, 2006) (Table 2).

Examples of mycobacterial lipoproteins

The 19 kDa antigen (LpqH, Rv3763) of *M. tuberculosis* (see Table 2 for summary of lipoproteins discussed) is a glycosylated lipoprotein. Early on, this protein was shown to induce a TLR-2-dependent bactericidal response in macrophages (Thoma-Uszynski *et al.*, 2001). Recently, the 19 kDa antigen was described as an adhesin, binding to the mannose receptor of THP-1 monocytic cells and thereby stimulating phagocytosis (Diaz-Silvestre *et al.*, 2005). The 19 kDa antigen was shown to induce interleukin-1 and -12, and tumour necrosis factor- α (TNF- α) through TLR2-signalling in macrophages. Prolonged exposure to the 19 kDa lipoprotein inhibits IFN- γ production and major histocompatibility (MHC) class II expression. These findings suggest that, at least in part, persistent TLR2 signalling enables *M. tuberculosis* to evade T cell responses and persist as a long-term infection. An *M. tuberculosis* 19 kDa knock-out mutant was reported to be slightly attenuated in IFN- γ activated monocyte derived macrophages (Stewart *et al.*, 2005).

Table 2. Distribution of lipoproteins in mycobacteria and sequence identities/similarities

Name	<i>M. tuberculosis</i> *	<i>M. leprae</i> †	<i>M. avium</i> ‡	<i>M. marinum</i> ‡	<i>M. smegmatis</i> §
19 kDa	LpqH	48/65	75/86	48/51	39/51
38 kDa	PstS1	33/48	36/48	32/46	34/49¶
PstS2	PstS2	62/77¶	77/85	66/77	45/56¶
PstS3	PstS3	77/86	78/86	77/83	48/61
LpqB	LpqB	87/92	86/92	82/87	72/81
LprF	LprF	30/48#	29/51#	65/72	32/48#
LprJ	LprJ	34/56	66/85	71/82	41/62
RpfB	RpfB	82/89	85/92	80/86	73/81
24kDa	LppX	76/85	34/52#	73/85	30/47#
LpqW	LpqW	79/85	81/88	72/77	74/82

**M. tuberculosis* H37Rv lipoproteins were used as a query.

†BLAST on http://www.ncbi.nlm.nih.gov/sutils/genom_table.cgi.

‡BLAST on http://www.sanger.ac.uk/cgi-bin/blast/submitblast/m_marinum.

§BLAST on <http://tigrblast.tigr.org/cmr-blast/>.

||Same ORF as identified by PstS2.

¶Same ORF as identified by PstS3.

#Same ORF as identified by LprG.

Phosphate transport receptors (Pst): three homologues of the periplasmic ABC phosphate-binding receptor PstS of *E. coli* have been described in *M. tuberculosis* [PstS1 (Rv0934), PstS2 (Rv0932c) and PstS3 (Rv0928)]. Expression of these proteins increases under phosphate-limiting conditions. *M. tuberculosis* mutants deficient in PstS1 and PstS2 showed decreased c.f.u. in lungs and spleens of mice, indicating a role in virulence (Peirs *et al.*, 2005). Vaccination of C57BL/6 mice with PstS3 DNA protected against challenge with *M. tuberculosis* (Romano *et al.*, 2006). Compared to PstS3 vaccination, vaccination with PstS2 and PstS1-DNA induced only modest reduction in c.f.u. counts.

LpqB (Rv3244c) is a particularly interesting lipoprotein, as it is one of the 233 conserved signature proteins of the actinobacteria (Gao *et al.*, 2006). *lpqB* is located immediately downstream of the two-component signal transduction system MtrAB. In this system MtrB is the transmembrane sensor-kinase and MtrA is the cytoplasmic response regulator. Lipoproteins have been shown to function as accessory proteins of sensor-kinase systems and it may be assumed that LpqB modulates signal sensing by MtrB (Hoskisson & Hutchings, 2006).

Lipoproteins LprF (Rv1368) and LprJ (Rv1690) exhibited protein-protein interactions with the histidine kinase KdpD in a two-hybrid screen. Activation of the Kdp signal transduction pathway appears to be the primary response to environmental osmotic stress in both *M. tuberculosis* and *M. smegmatis*. The histidine kinase domain of Kdp has been suggested to form ternary complexes with LprF and LprJ and it was speculated that these proteins function as ligand-binding proteins. Co-induction of LprJ with a cluster of genes involved in cell wall integrity suggests that LprJ is

involved in this process (Boshoff *et al.*, 2004). Alternatively, LprF or LprJ could function as accessory proteins as discussed for LpqB.

RpfB (Rv1009) is the only lipoprotein among the five resuscitation-promoting factor (Rpf) proteins of *M. tuberculosis*. Rpf proteins stimulate dormant cells to divide (Keep *et al.*, 2006). They have structural homology to glycoside hydrolases and cleave peptidoglycan; however, the exact mechanism by which these proteins promote resuscitation remains elusive. Investigations in a mouse model of *M. tuberculosis* persistence and reactivation indicated that inactivation of *rpfB*, but not inactivation of *rpfA*, C, D or E, delayed reactivation, suggesting a unique role of RpfB in resuscitation (Tufariello *et al.*, 2006). Whether this unique role of RpfB is related to the lipid anchor remains to be determined.

Investigation of LpqW and LppX revealed that these lipoproteins are key players in synthesis and transport of the unique components of the mycobacterial cell envelope. LppX (Rv2945c) is a lipoprotein involved in translocation of complex lipids, the phthiocerol dimycocerosates (DIM), to the outer membrane. Structural elucidation of LppX revealed the presence of a hydrophobic cavity suitable for binding the large lipophilic side chain of DIM (Sulzenbacher *et al.*, 2006). Orthologues of LppX are only present in mycobacteria which synthesize DIM (e.g. *M. tuberculosis*), and are absent from DIM-negative mycobacteria (e.g. *M. smegmatis* or *M. avium*). An *M. tuberculosis* mutant deficient in LppX is attenuated in a mouse model of infection. Attenuation is associated with a failure to release DIM into the culture supernatant rather than reduced DIM synthesis (Sulzenbacher *et al.*, 2006).

LpqW (Rv1166) is a highly conserved, essential lipoprotein involved in the synthesis of cell wall components. Structural analyses suggest that LpqW is derived from substrate-binding proteins, which in mycobacteria and other micro-organisms of the subfamily *Corynebacterinae* (also producing LAM) has evolved to match the specific needs in the synthesis of the cell wall components phosphatidyl-*myo*-inositol mannoside (PIM) and LAM. LpqW acts at the branching point of the PIM/LAM pathway and converts the last common intermediate, the tetramannosylated phosphatidylinositol mannoside PIM₄, into the LAM pathway (Kovacevic *et al.*, 2006; Marland *et al.*, 2006).

Conclusions

Lipidation of proteins is required for their anchoring and sorting to the cellular surface. Recent investigations point to the importance of lipoprotein-synthesizing enzymes as well as individual lipoproteins in the biology of mycobacteria. Mycobacterial lipoproteins are crucial for synthesizing the unique mycobacterial cell envelope, sensing of and protection from environmental stress and participation in host-pathogen interaction. Due to their contribution to virulence, lipoproteins and the enzymes of the lipoprotein synthesis pathway represent promising drug targets. In addition some lipoproteins confer a protective immune response and thus may qualify as subunit vaccines. Despite progress in lipoprotein research much remains to be learned with respect to the synthesis, localization and function of mycobacterial lipoproteins and their role in host-pathogen interaction.

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