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List. monocytogenes' peptization. meso-diaminopimelic acid residues are amidated AsnB which also has impact bacteria Surface characteristics host cell invasion.

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Abstract

It was discovered that a Listeria monocytogenes ScottA mutant with a transposon in the asnB gene's 5' untranslated region was extremely sensitive to the antibiotic t-cinnamaldehyde. Here, we describe the functional characterization of AsnB in intracellular infection and peptidoglycan (PG) modification. Although sequence alignment revealed that AsnB of Listeria is closely linked to a collection of homologs that catalyse the amidation of meso-diaminopimelic acid (mDAP) residues in the peptidoglycan of other bacterial species, this protein is designated as a glutamine-dependent asparagine synthase. According to a structural study of the peptidoglycan from an asnB mutant compared to isogenic wild-type (WT) and complemented mutant strains, asnB mediates mDAP amidation in L. monocytogenes. Numerous peptidoglycan and cell surface-related abnormalities were brought on by mDAP amidation deficiency in asnB. It was discovered that a Listeria monocytogenes ScottA mutant with a transposon in the asnB gene's 5' untranslated region was extremely sensitive to the antibiotic t-cinnamaldehyde. Here, we describe the functional characterization of AsnB in intracellular infection and peptidoglycan (PG) modification. Although sequence alignment revealed that AsnB of Listeria is closely linked to a collection of homologs that catalyse the amidation of meso-diaminopimelic acid (mDAP) residues in the peptidoglycan of other bacterial species, this protein is designated as a glutamine-dependent asparagine synthase. According to a structural study of the peptidoglycan from an asnB mutant compared to isogenic wild-type (WT) and complemented mutant strains, asnB mediates mDAP amidation in L. monocytogenes. Numerous peptidoglycan and cell surface-related abnormalities in asnB, bacterial virulence, and InlA were caused by mDAP amidation deficiency.

***Keywords:** Peptidoglycan, biotechnology, biological*

Introduction

Bacterial cells are enclosed by a stiff peptidoglycan (PG) cell wall, whose major universal purpose is to preserve cell integrity and form, particularly in hypo-osmotic settings when cell lysis would otherwise be advantageous (Vollmer et al., 2008). The mesh-like structure of PG, which is made up of long polymeric chains of N-acetyl glucosamine (GlcNAc) and N-acetylmuramic acid (MurNAc) heterodisaccharides and is cross-linked by peptide side chains, provides the tensile strength needed for this function. Although the fundamental structure has been effectively preserved, the PG chemical structures in various bacteria vary greatly. This variation results from the presence or absence of modifying substituents on the sugar and amino acids, as well as changes in the composition of the amino acids and side chain cross-linking. units of acid. Different PG structures have also been seen in various bacteria, depending on their developmental stage and environmental conditions. In spore-forming bacteria, a distinctive and singular-lactam PG modification is present in the so-called spore cortex, a thick protective PG layer surrounding the germ cell wall that does not have this modification. This enables particular hydrolases placed on the spore surface to specifically break the cortex upon spore germination without harming the germ cell wall.

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PG serves as a scaffold for surface proteins with a variety of roles in addition to its structural function. PG or its fragments also operate as a molecular signal in a number of symbiotic or pathogenic interactions with animal or plant hosts. The inclusion of several special or uncommon building components, such as MurNAc, meso-diaminopimelic acid (mDAP), and D-amino acids, as well as the significant structural variation in PG, which is produced in part by enzymatic changes, provides the high specificity necessary for such signalling. Both gram-positive and gram-negative bacteria frequently undergo PG changes, which are frequently crucial for pathogen pathogenicity. For instance, pathogens such as different streptococci and Listeria monocytogenes undergo N-deacetylation of GlcNAc, which inhibits PG hydrolysis by lysozyme and aids cells in avoiding the immunological reaction when under infection. O-acetylation of MurNAc is likely the most common and best-studied alteration, and it has also been linked to lysozyme resistance and virulence in pathogens including Neisseria gonorrhoeae and Staphylococcus aureus. In comparison to the sugar backbone, the stem peptide has even more variants. The most common and well-researched examples of these include amidation of D-iso-Glu and mDAP residues on their respective -carboxyl and -carboxyl groups. These involve the addition of substituents. While many genera and species of gram-positive bacteria undergo these amidation alterations, the only example of this in gram-negative bacteria that has been documented thus far is the amidation of the carboxyl group of mDAP in Acetobacteraceae (Esp).

Numerous bacterial characteristics and functions have been discovered to be impacted by stem peptide amidation. MurT/GatD mediates the amidation of D-iso-Glu, which is crucial for both S. aureus and Streptococcus pneumoniae. Reduced expression of the murT/gatD operon increased S. aureus' sensitivity to -lactam antibiotics and lysozyme. An enzyme that is similar to the glutamine-dependent asparagine (Asn) synthetase present in numerous species, such as AsnB of Escherichia coli, mediates the carboxyl amidation of mDAP. The amidotransfer from glutamine to aspartate for the production of asn as well as to mDAP for the modification of the PG stem peptide can be catalysed by at least some of these promiscuous enzymes. Three homologs, AsnB, AsnO, and AsnH, are encoded by Bacillus subtilis, and while none of them is necessary for Asn synthesis in B. subtilis, as even a triple knockout mutant may still grow without Asn, albeit at a slower rate. However, each of them can supplement the Asn shortfall of an E. coli Asn auxotroph. Later research revealed that AsnB mediates mDAP amidation, whereas neither of its two homologs.