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Peptidoglycan–outer membrane attachment generates periplasmic pressure to prevent lysis in Gram–negative bacteria

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Abstract

Bacteria are subject to a substantial concentration differential of osmolytes between the interior and exterior of the cell, resulting in turgor pressure. Failure to mechanically balance this turgor pressure causes cells to burst. Here, using microfluidics, imaging, biochemistry and mathematical modelling, we analysed how *Escherichia coli* cells with structural mutations in the envelope respond to hypoosmotic shocks. We show that the peptidoglycan cell wall forms a mechanical unit with the outer membrane that limits periplasmic volume increase under hypoosmotic shock, allowing osmotic pressure build-up in the periplasm. In turn, this periplasmic pressure balances cytoplasmic turgor across the inner membrane, preventing cell lysis and death. Thus, while the peptidoglycan layer is necessary, it is not sufficient to maintain turgor and protect cells from lysis. We propose a model in which the entire cell envelope, including the periplasm, collectively enables Gram-negative bacteria to overcome osmotic challenges.