

CELLULAR MICROBIOLOGY

Bacterial pea shooters



a bacterial surface structure that may enable long-distance delivery of outer membrane vesicles



Gram-negative bacteria produce outer membrane vesicles with roles in processes such as communication, pathogenesis and nutrition. Outer membrane vesicles can diffuse across long distances in environments with abundant water. However, in environments with limited water availability, such as soil, relying on diffusion for the spread of these

vesicles would be ineffective. Now, Hickey and colleagues describe a bacterial surface structure that may enable long-distance delivery of outer membrane vesicles in a diffusion-independent manner.

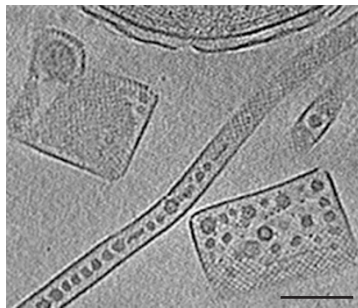
The authors studied cultures of a soil betaproteobacterium from the family Comamonadaceae, *Delftia* sp. Cs1-4, by transmission electron microscopy (TEM) and electron cryotomography, and found tubular structures (>6 µm long) that projected from the cell surfaces. These structures, which the authors call nanopods, are crystalline-like tubes that enclose chains of vesicle-like structures and are contiguous with the proteinaceous surface layer (S layer) covering the cells. Mass spectrometry of nanopods revealed a high abundance of a protein of unknown function (designated nanopod protein A (NpdA)), together with various putative outer membrane proteins, periplasmic enzymes and secreted proteins. The nanopod vesicles were found to contain lipopolysaccharide, further supporting the idea that

they are bona fide outer membrane vesicles.

Immunogold-labelling experiments showed that NpdA is localized to nanopods and the cell envelope. Deletion of *npdA* resulted in a strain that lacked nanopods and the S layer but still produced outer membrane vesicles. Thus, NpdA is a surface-associated protein that is required for the formation of both the S layer and nanopods. Interestingly, nanopod formation was confirmed by TEM in closely related bacteria that are from the same family and also have *npdA*-like genes.

Nanopods may allow some soil bacteria to use outer membrane vesicles for long-distance projection of metabolic activity. However, some of the identified nanopod-producing organisms are plant pathogens or animal symbionts, suggesting that nanopods are tailored for unique functions in other habitats.

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A slice through a three-dimensional electron cryotomographic reconstruction of a culture of *Delftia* sp. Cs1-4, showing nanopods in linear form as they grow attached to cells, and the square-shaped structures formed by detached nanopods. The crystalline outer surface appears as a cross-hatched area, and outer membrane vesicles are circular elements. The scale bar represents 100 nm. Image courtesy of W. J. Hickey, University of Wisconsin–Madison, USA, and G. J. Jensen, California Institute of Technology, Pasadena, USA.

ORIGINAL RESEARCH PAPER Shetty, A. et al. Nanopods: a new bacterial structure and mechanism for deployment of outer membrane vesicles. *PLoS ONE* 6, e20725 (2011)