REPLY TO DESIKAN ET AL.:

Micelle formation among various mechanisms of toxin pore formation

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Pore-forming toxins (PFTs) are a diverse class of membrane-active proteins employed primarily by bacteria for unregulated perforation of lipid membranes (1). Based on molecular dynamics (MD) simulations (2), electron cryomicroscopy (cryoEM) structures (3), and atomic force microscopy (AFM) experiments (4), we recently identified two distinct pathways for lipid efflux from large pores (diameter >30 nm) formed by the β-PFT pneumolysin (PLY). Lipids leave the PLY ring laterally upon slow membrane insertion of the β-strands forming the pore-lining β-barrel; by contrast, upon fast insertion, the membrane within the ring bends into a small vesicle that then gets expelled from the pore (2). In their letter, Desikan et al. (5) describe a variant of the vesiculation pathway, in which the lipids trapped inside a small α-PFT ring (diameter <10 nm) form a micelle, which then leaves the ring.

Desikan et al. (5) performed MD simulations of ClyA as an example for a small cytotoxin. In their simulation setup, the fully formed ClyA ring traps the lipids inside the pore. By contrast, their earlier simulations have shown that slow assembly of membrane-inserted protomers allows the lipids to leave laterally (6), reminiscent of the lateral-escape pathway seen in our PLY simulations (2). Alternatively, the crystal structure of ClyA suggests that in the case that a fully formed ring is inserted, the lipids of the small plug would be wedged out rather than cut out (7). Lateral escape (6), wedging (7), and micelle formation (5) provide alternative lipid efflux pathways from small PFT pores.

From a mechanistic perspective, pore opening via micelle formation (5) is the small-pore variant of the vesiculation mechanism that we propose in our recent paper (2). Both in narrow ClyA rings and in wide PLY rings, a fully cut-out patch of lipids reshapes into a form that minimizes the edge tension and lets the lipids escape from the ring vertically instead of laterally from a partially formed pore. AFM studies of various large pores have shown that the exact mechanism depends on the pore-forming protein, its oligomeric state (arc, slit, or ring), the membrane composition, and the environmental conditions (4, 8–10).

In summary, there are two principal pathways of lipid removal: vertical expulsion [by vesiculation (2, 8) or micelle formation (5)] and lateral retreat [by lipid outflow (2, 6) or wedging (7)], driven by lipid repulsion from the hydrophilic inner wall of the pore (2). PFTs may have evolved this variability in the pore formation pathway to combat cellular defense mechanisms.

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The authors declare no competing interest.

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