Nature Reviews Gastroenterology & Hepatology | Published online 23 Aug 2017; doi:10.1038/nrgastro.2017.118

A new way out for lysozyme

> the secretion of lysozyme through the autophagy pathway during *S.* Typhimurium infection is triggered by ER stress

The intestinal epithelium contains Paneth cells that protect against bacterial invasion by secreting antimicrobial proteins such as lysozyme. However, infection with pathogenic microorganisms can trigger endoplasmic reticulum (ER) stress in Paneth cells and compromise their secretion of antimicrobials. Bel and colleagues now show that lysozyme is released by Paneth cells through an alternative autophagy-based secretion pathway during infection with pathogenic bacteria.

Salmonella enterica serovar Typhimurium (*S.* Typhimurium) can invade cells of the epithelium and triggers ER stress and autophagy. In mice that were infected with *S.* Typhimurium, immunofluorescence, transmission electron microscopy and co-immunoprecipitation assays showed that Paneth cells contained an elevated number of lysozyme-positive vesicles, with the typical characteristics of autophagosomes (double-membrane bound, microtubule-associated protein 1 light chain 3-positive ($LC3^+$)). These vesicles did not fuse with lysosomes, colocalized with a marker of the secretory autophagy pathway (Rab8α) and some $LC3^+$lysozyme$^+$ vesicles fused with the apical surface of Paneth cells and secreted lysozyme into the intestinal lumen. These findings suggest that *S.* Typhimurium infection of mice induces an alternative autophagy-related secretory pathway, resulting in lysozyme secretion.

The relative importance of conventional and autophagy-dependent secretion pathways was tested when *ex vivo* Paneth cell crypts were infected with *S.* Typhimurium and treated with inhibitors. Lysozyme secretion was not affected when ER–Golgi trafficking or lysosome acidification were inhibited. By contrast, inhibiting autophagosome nucleation decreased lysozyme secretion, and secretions from these crypts could not kill bacteria. To identify a trigger for secretory autophagy, the ER stress response in *S.* Typhimurium-infected mice was inhibited with tauroursodeoxycholic acid (TUDCA), which inhibited the secretory autophagy of lysozyme. By contrast, inducing ER stress increased secretory autophagy of lysozyme. Importantly, inhibiting secretory autophagy of lysozyme with TUDCA increased the disease burden of *S.* Typhimurium-infected mice; an effect that could be rescued by lysozyme gavage. These data suggest that the secretion of lysozyme through the autophagy pathway during *S.* Typhimurium infection is triggered by ER stress.

In the intestinal epithelium, the activation of antibacterial autophagy requires the expression of the Toll-like receptor (TLR) signalling adaptor MYD88. The authors found that MYD88 expression was essential in dendritic cells (DCs) for secretory autophagy of lysozyme in Paneth cells. Interestingly, treating MYD88-deficient mice with interleukin-22 (IL-22) rescued lysozyme secretion. This suggests a role for a known pathway where a bacterial signal is sensed by DCs and relayed to epithelial cells via the downstream activation of type 3 innate lymphoid cells (ILC3s) and secretion of IL-22. In support of this, *Rorc*$^{-/-}$ mice (deficient in ILC3s and T helper 17 cells) also had disrupted secretory autophagy. The authors suggest that this circuit may provide a signal that allows Paneth cells to rapidly activate secretory autophagy upon experiencing ER stress.

Finally, the authors found that lysozyme secretion and bacterial killing were reduced in the crypts of *S.* Typhimurium-infected mice with mutations in the autophagy related 16-like 1 gene (*Atg16L1*). These findings could help explain why mutations in this gene are a risk factor for Crohn's disease in humans.

Shimona Starling, Cross-Journal Associate Editor

This article originally appeared in *Nat. Rev. Immunol.* (doi:10.1038/nri.2017.98).

ORIGINAL ARTICLE Bel, S. *et al.* Paneth cells secrete lysozyme via secretory autophagy during bacterial infection of the intestine. *Science* http://dx.doi.org/10.1126/science.aal4677 (2017)