Featured Publication Note
Professor Peter Cullen and colleagues
Department of Biochemistry, University of Bristol

The role of sorting nexin-1 (SNX1) during *Salmonella* infection

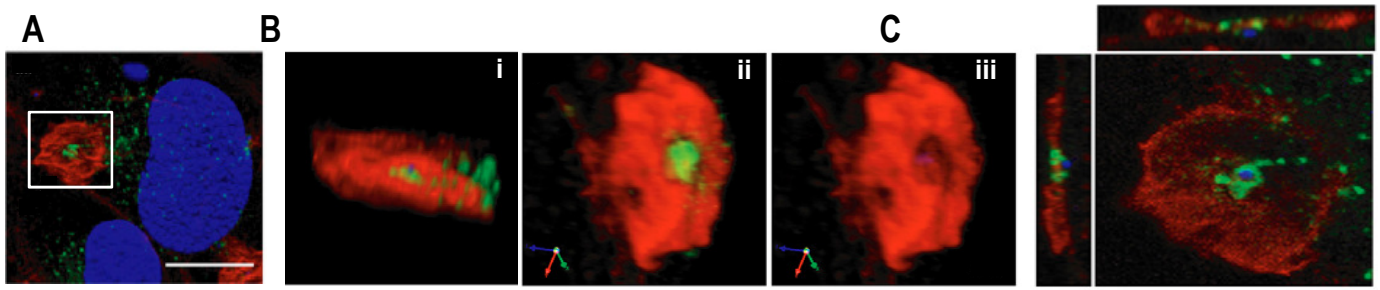

Figure 1: The boxed region in part A shows that SNX1 (green) accumulates at membrane ruffles (Scale bar: 10 µm). A magnification of this boxed region is shown in part B, in which different orientations of the 3D rendered view are displayed, with the green (SNX1) channel omitted in B(iii). Part C shows a single optical *z*-section of the maximum projections shown in figures A and B, with insets providing the respective *YZ*- and *XZ*-views.

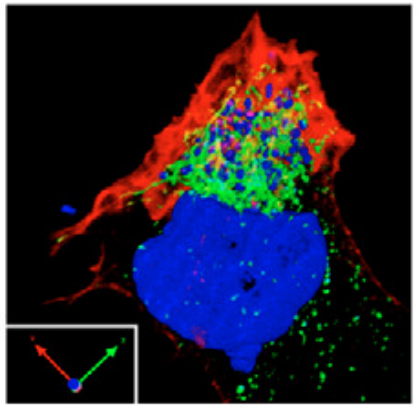

Figure 2: MDCK cells containing a GFP-SNX1 fusion protein (green) were fixed after a 15 minute infection with SL1344, and treated with TRITC-phalloidin (red) and DAPI (blue). Images of a deconvolved stack are shown. GFP-SNX1 creates a meshwork around bacteria.

Professor Peter Cullen and colleagues are interested in the role of the mammalian sorting nexins in host-pathogen interactions. *Salmonella enterica* serovar Typhimurium (*S.* Typhimurium) is the leading cause of human gastroenteritis and invades non-phagocytic cells by actively inducing its uptake. Once inside, *S.* Typhimurium establishes a replication niche within membranous *Salmonella*-containing vacuoles (SCVs) which are enriched in phosphatidylinositol 3-monophosphate [PtdIns(3)*P*] and 'blend in' with the host's endosomal system.

In this work the researchers use a combination of live cell and immunofluorescence microscopy (using the PerkinElmer Ultra*VIEW* LCI confocal Optical Scanner) to show that sorting nexin-1 (SNX1) is crucially involved in remodeling nascent SCVs. SNX1 contains a sorting nexin phox (PX)-homology domain that binds to PtdIns(3)*P* and a membrane-binding module (called a BAR domain) which induces membrane tubulation.

MDCK cells were infected with *S.* Typhimurium strain SL1344 for 15 minutes, then fixed, immunolabeled with anti-SNX1 (green), and treated with TRITC-phalloidin (red) and DAPI (blue). Ten optical *z*-slices were acquired (z=5.37 µm) and data sets were deconvolved, 3D rendered and animated using Volocity software (Figure 1; see also http://jcs.biologists.org/content/vol121/issue12/images/data/2027/DC1/018432-Movie1.mov). SNX1 was shown to be readily recruited to sites of *Salmonella* invasion, and was particularly found clustered underneath actin-rich membrane ruffles. This recruitment of SNX1 was found to induce the formation of numerous long-range tubules. In order to better visualize the full extent of tubulation, cells expressing GFP-SNX1 were infected with SL1344, fixed and treated with TRITC-phalloidin and DAPI. Confocal imaging was used to acquire 29 optical *z*-slices (z=4.76 µm) and data sets were deconvolved, 3D rendered, animated and exported using Volocity software (Figure 2; see also http://jcs.biologists.org/content/vol121/issue12/images/data/2027/DC1/018432-Movie3.mov). The results revealed an extensive tubular meshwork of GFP-SNX1 around the invading bacteria 15 minutes after infection.

The authors went on to show that SNX1-induced tubulation is required for SCV biogenesis as suppression of SNX1 leads to defects in the intracellular progression and replication of the invaded *Salmonella*.

Figures adapted with permission from the Journal of Cell Science: Bujny M V, Ewels P A, Humphrey S, Attar N, Jepson M A and Cullen P J (2008). *Journal of Cell Science*. 121 (12): 2027-2036 (http://jcs.biologists.org/cgi/content/full/121/12/2027).