

Description of Additional Supplementary Files

File Name: Supplementary Movie 1

Description: **3D STED animation.** 3D animated reconstruction of two podosomes from STED micrographs of macrophage stained for LSP1 (green) and F-actin (red), as shown in Fig. 1F.

File Name: Supplementary Movie 2

Description: **Podosome cluster dynamics, control cell.** Live imaging of macrophages targeted with control siRNA and expressing pLifeact-tagRFP (gray scale) to visualize F-actin, as shown in Fig. 2A. Experiment performed with UltraVIEW VoX system (Perkin Elmer) spinning disk microscope, at 37° C and controlled atmosphere. Frames were acquired every 60 sec for 1h. Scale bar = 10 μ m

File Name: Supplementary Movie 3

Description: **Podosome cluster dynamics, LSP1 knockdown cell (siRNA #1).** Live imaging of macrophage targeted with LSP1 siRNA #1 and expressing pLifeact-tagRFP (gray scale) to visualize F-actin, as shown in Fig. 2B. Experiment performed with UltraVIEW VoX system (Perkin Elmer) spinning disk microscope, at 37° C and controlled atmosphere. Frames were acquired every 60 sec for 1h. Scale bar = 10 μ m

File Name: Supplementary Movie 4

Description: **Podosome cluster dynamics, LSP1 knockdown cell (siRNA #2).** Live imaging of macrophage targeted with LSP1 siRNA #2 and expressing pLifeact-tagRFP (gray scale) to visualize F-actin, as shown in Fig. 2C. Experiment performed with UltraVIEW VoX system (Perkin Elmer) spinning disk microscope, at 37° C and controlled atmosphere. Frames were acquired every 60 sec for 1h. Scale bar = 10 μ m

File Name: Supplementary Movie 5

Description: **Localization of LSP1 and supervillin in macrophage changing direction.** Live imaging of macrophage overexpressing GFP-LSP1 and supervillin-RFP, as shown in Suppl. Fig. 7A. Experiment performed with UltraVIEW VoX system (Perkin Elmer) spinning disk microscope, at 37° C and controlled atmosphere. Frames were acquired every 35 sec. for 35 min. Scale bar = 10 μ m

File Name: Supplementary Data 1

Description: **Values for podosome and cell analysis.**