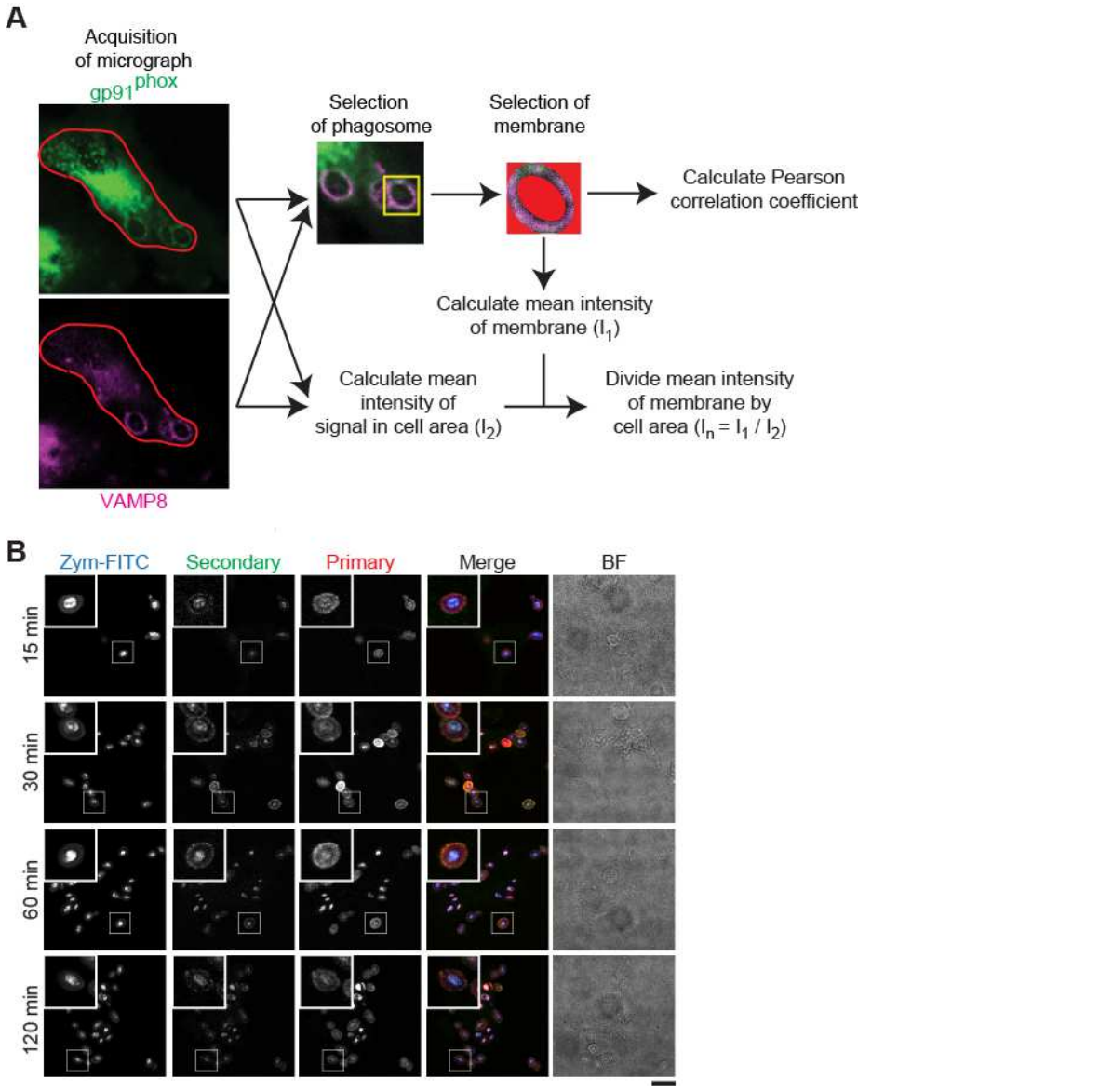
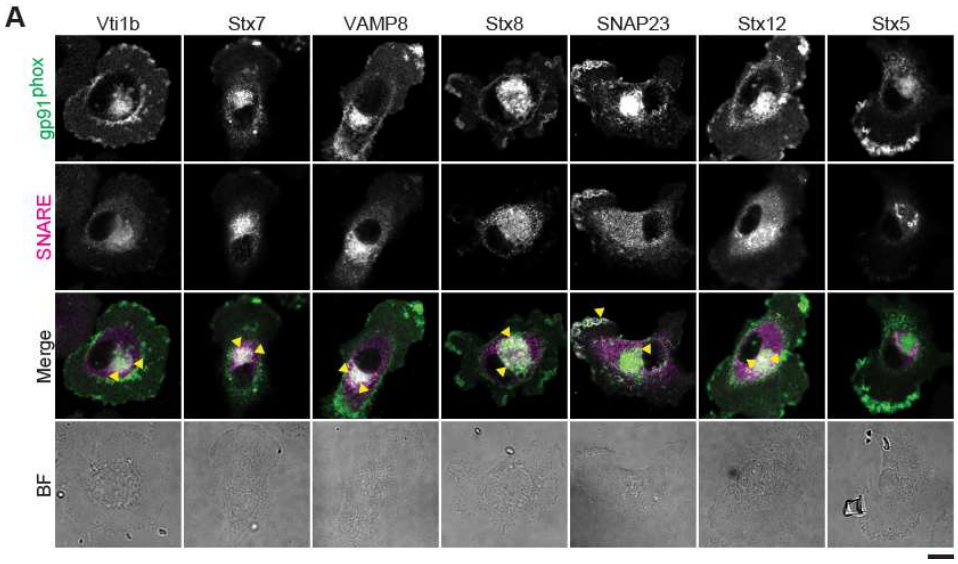


Fig. S1: Calculation of the Pearson correlation coefficients on the phagosomes and the phagosomal enrichment factors, and control experiment for phagosomal sequestration of gp91^{phox}. A) Example confocal image of a dendritic cell immunostained for gp91^{phox} (green) and VAMP8 (magenta). For the Pearson correlation coefficients, the membranes of individual phagosomes were manually selected based on morphology (excluded area masked in red) and the correlation between the channels was calculated. For the normalized intensity values (I_n), the membranes of individual phagosomes were also manually selected and the mean fluorescence intensitity (I_1) was determined for each phagosome. These values were then divided by the mean fluorescence intensity over the entire imaged cell area (I_2) to correct for different staining efficiencies and expression levels among cells/donors (perimeter of the imaged cell area depicted in red). The phagosomal enrichment is defined as the percentage of phagosomes with normalized intensity values >1. **B)** Representative confocal images of dendritic cells stimulated with zymosan conjugated with FITC (Zym-FITC; blue) and labeled with an Alexa fluor 647-labeled antibody raised against FITC (primary; red). After 1 hour of stimulation, cells were immunostained with an Alexa fluor 568-labeled secondary antibody (secondary; green). The images were quantified from the Alexa fluor 568 signal relative to the Alexa fluor 647 signal (shown in main figure 1E). BF: bright field. Insets: magnification of zymosan-containing phagosomes. Scale bar, 10 µm.



B

	w/o zymosan	+ zymosan
gp91	0.89	0.92
Vti1b	0.69	0.71
Stx7	0.64	0.60
VAMP8	0.68	0.70
Stx8	0.57	0.61
SNAP23	0.68	0.60
Stx12	0.60	0.53
Stx5	0.41	0.43

Fig. S2: Gp91^{phox} colocalizes with SNAREs in resting cells. A) Confocal images of dendritic cells (without zymosan) immunostained for gp91^{phox} (green in merge) with Vti1b, stx7, VAMP8, stx8, SNAP23, stx12 or stx5 (magenta). Yellow arrowheads: cellular regions positive for both gp91^{phox} and the indicated SNARE. BF: bright field. See main figure 3B–C for quantification. Scale bar, 10 µm. **B)** The mean Pearson correlation coefficients of unstimulated and zymosan-pulsed cells of main figure 3B.

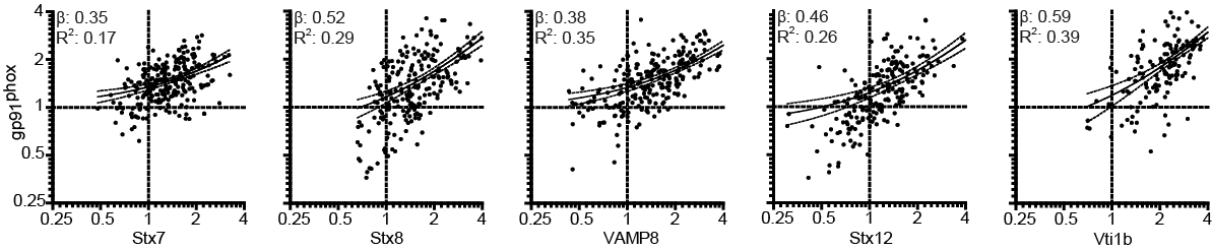


Fig. S3: Regression analysis of normalized intensity values on phagosomes. Similar to figure 3D. Normalized intensity values of $gp91^{phox}$ as a function of normalized intensity values of the indicated SNAREs for individual phagosomes (Log2-scale; solid lines: linear regression with 95% confidence intervals; β: regression coefficients; R^2: R-squared values).

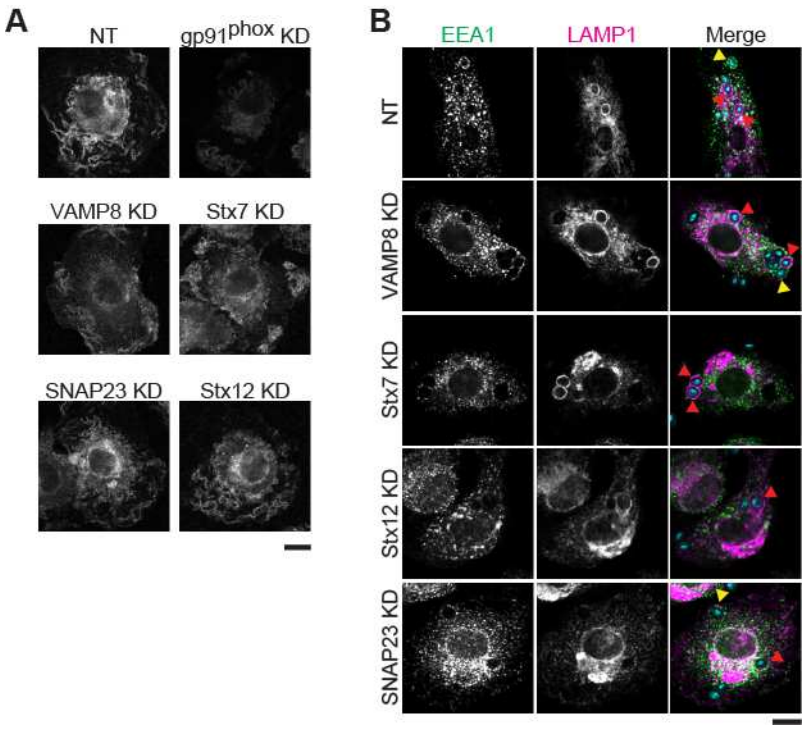


Fig. S4: Subcellular localization of NOX2, EEA1 and LAMP1 after knockdown. A) Representative confocal images of dendritic cells with knockdown of gp91^{phox}, VAMP8, stx7, SNAP23 and stx12 (without zymosan) immunostained for gp91^{phox}. **B)** Representative confocal images of dendritic cells with knockdown of gp91^{phox}, VAMP8, stx7, SNAP23 and stx12 stimulated with zymosan and immunostained for EEA1 (green in merge) and LAMP1 (magenta). Yellow arrowheads: phagosomes enriched for EEA1; red arrowheads: phagosomes enriched for LAMP1. Scale bars, 10 µm.