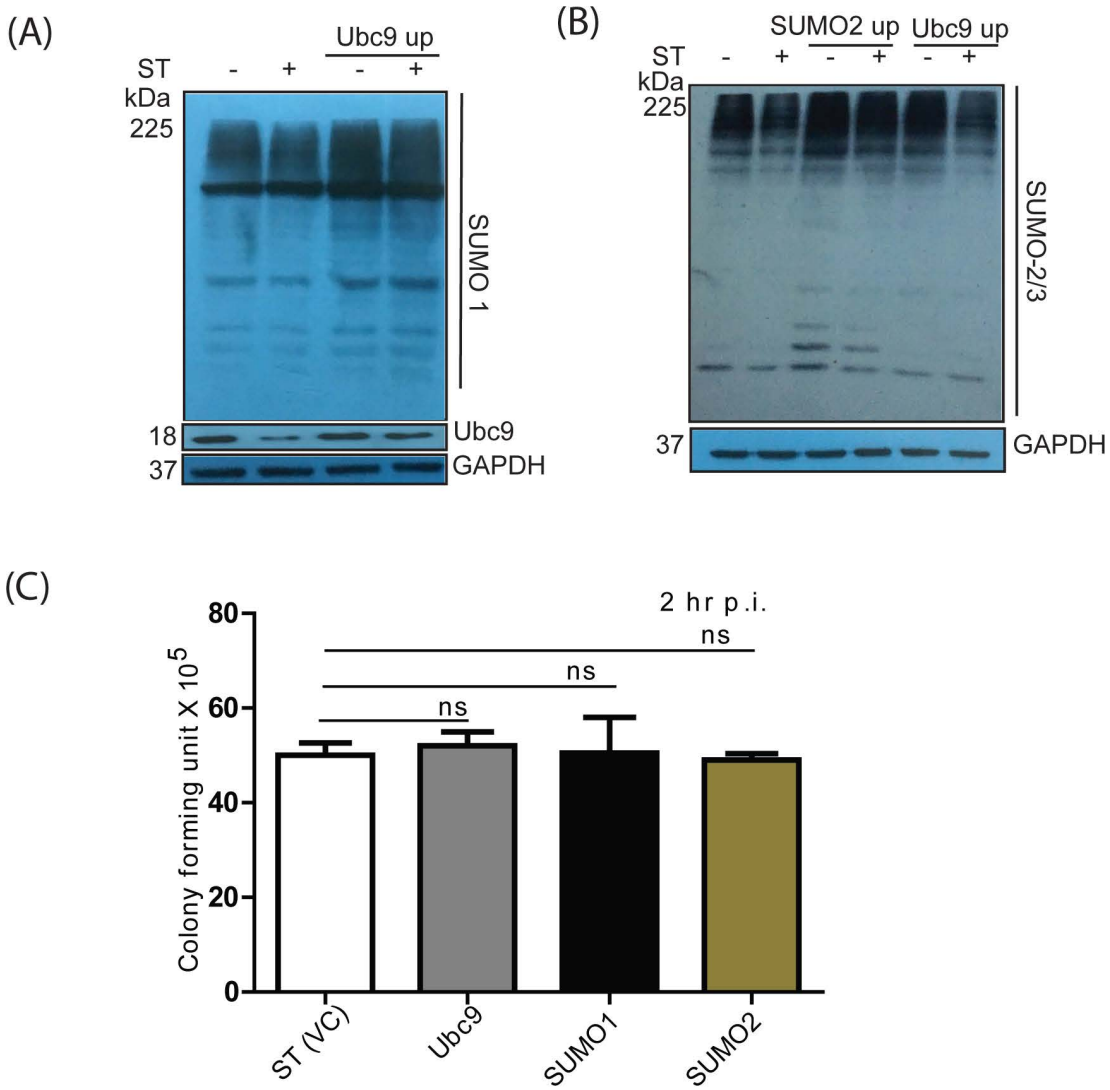


Fig. S1. (A) HCT8 cells were transfected using Lipofectamine 2000 with pcDNA Ubc9 plasmid DNA. Lysates were run on SDS-PAGE followed by immunoblotting for SUMO1, Ubc9 and GAPDH. (B) HCT8 cells were transfected with pcDNA Ubc9 or SUMO2/3 followed by ST infection and immunoblotting for SUMO 2/3. The lower blot represents the corresponding GAPDH. (C) Gentamicin protection assays were performed in HCT8 cells transfected with vector control plasmids or those encoding SUMO1, SUMO2/3 or Ubc9 and infected with ST for 2 hr. The colony forming units (CFU) were scored and plotted for the indicated sample. Mean ± SEM from three independent experiments was included in the plot. (D) Confocal microscopic images of HeLa cells transfected with pcDNA Ubc9 plasmid followed by infection with ST expressing mCherry for 7 hr and immunostained for LAMP1 to visualize *Salmonella* induced filaments (SIFs). Bar - 10μM (E) Quantitative representation of percentage of SIFs from three independent experiments where Mean ± SEM was plotted as described above. Statistical analysis was carried out using Student's t test. The sign "ns" indicates non-significant and "***" indicates a P value of ≤ 0.001.

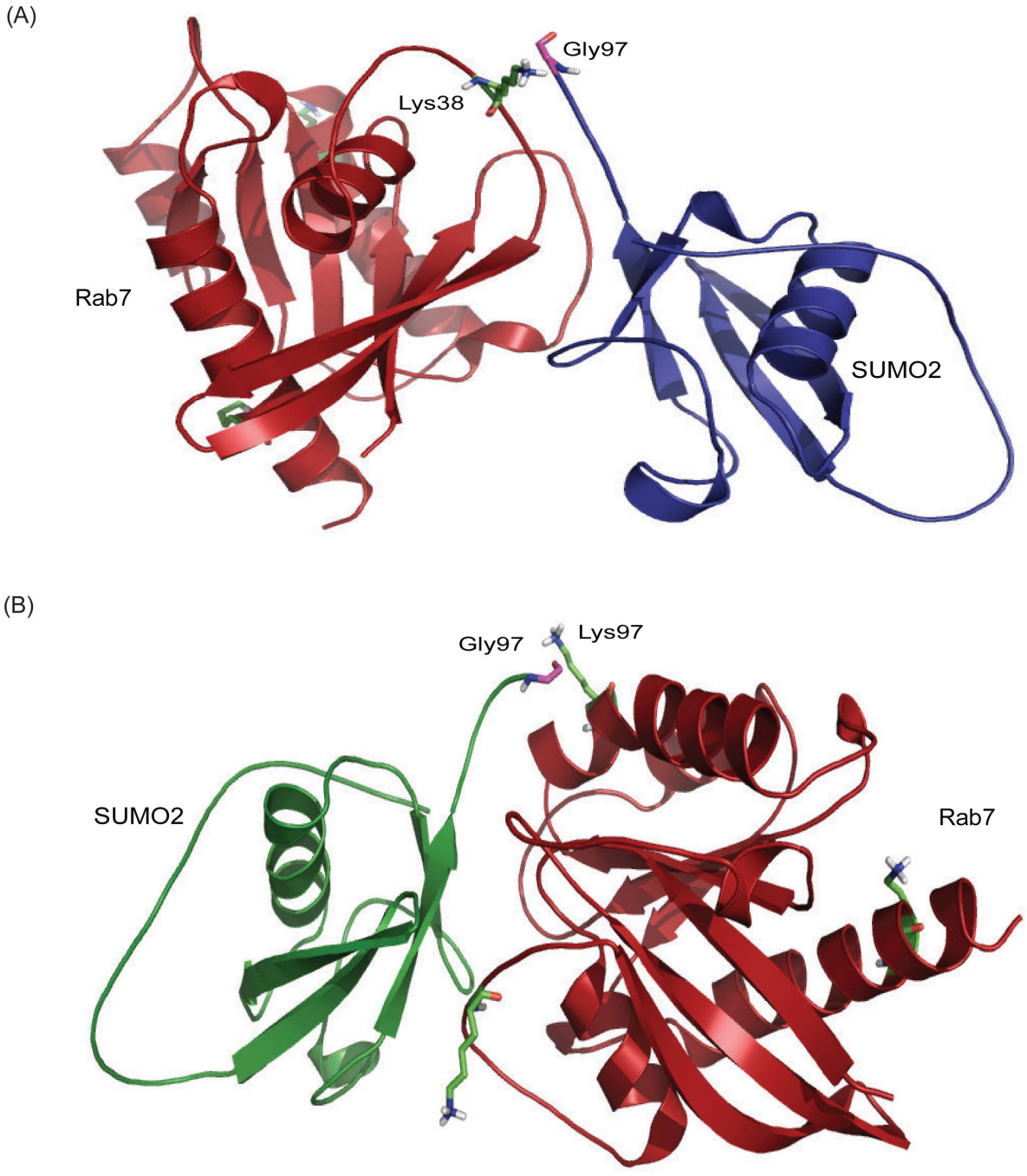


Fig. S2. Representative image obtained from Molecular Docking depicting complex of SUMO2-Rab7(A) Ribbon representation of docked model of SUMO2 (blue) in complex with Rab7 (red) at Lysine 38 (B) and Lysine 97 where SUMO2 is green and Rab7 is red.

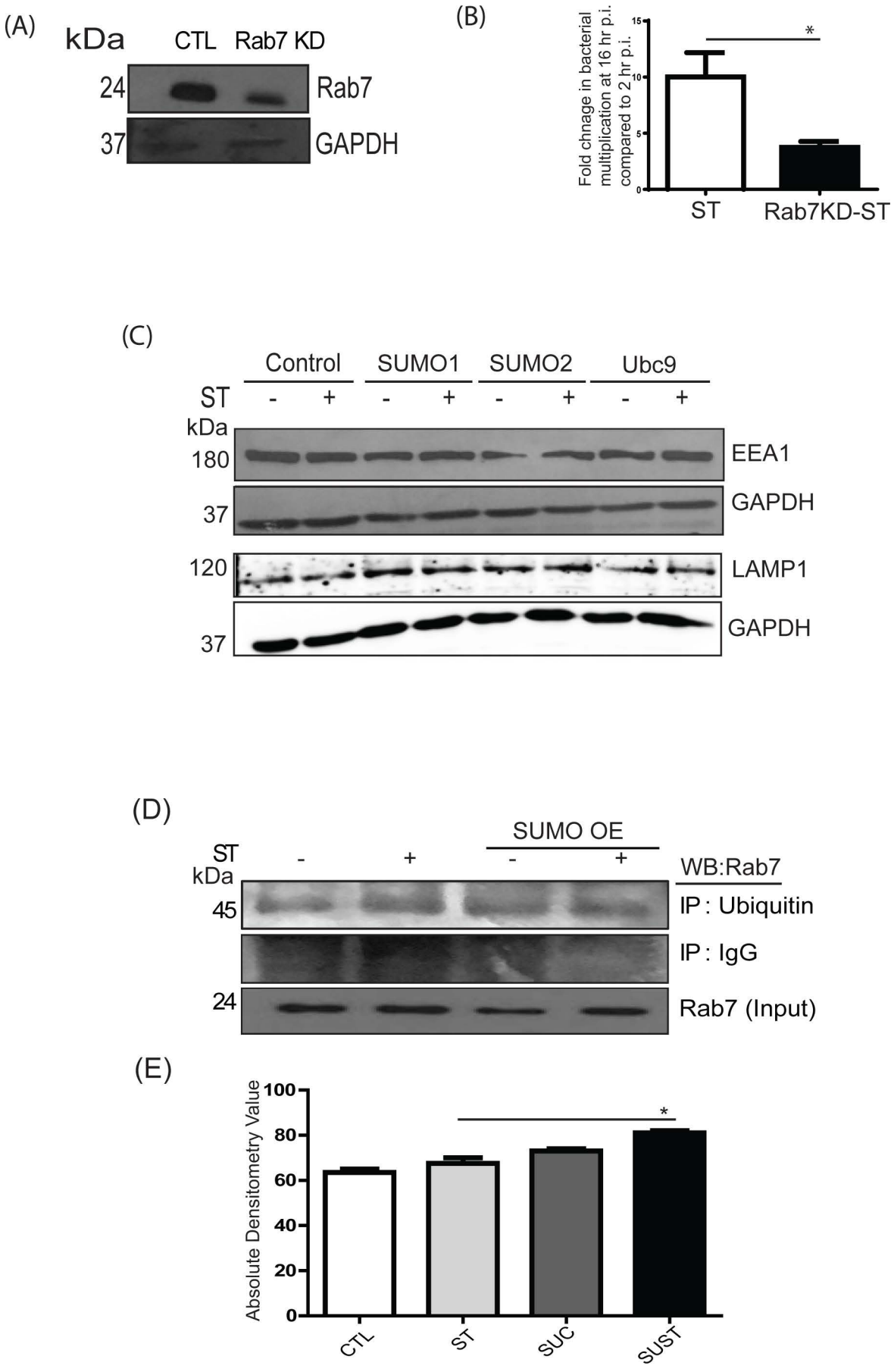


Fig. S3. Effect of on endocytic markers during ST infection. (A) HCT8 cells were transfected with scrambled siRNA in control sample and siRNA specific for Rab7 followed by immunoblotting showing the knock-down of Rab7. GAPDH was used as a loading control. (B) Gentamicin protection assay was performed in the HCT8 cells (similar to those shown in A) and infected with ST for 2 hrs or16 hrs. Relative fold change in bacterial multiplication at 16 hrs compared to 2 hrs was plotted. Mean ± SEM from three independent experiments was included in the plot. Statistical analysis was carried out using Unpaired Student's t test. The sign "*" indicates a P value of ≤ 0.05. (C)Immunoblot representing the expression of EEA1 (15 mins p.i.) and LAMP1 (4 hr p.i.) when cells were transfected with SUMO1, SUMO2, and Ubc9 constructs as mentioned above. GAPDH was used as a loading control. (D) Lysates of HCT 8 cell transfected with pcDNA Ubc9 or untreated cells that were infected with ST were immunoprecipitated (IP) using anti-Ubiquitin antibodies and probed with Rab7 antibodies or isotype control IgG antibodies. The corresponding densitometry values were plotted (E).

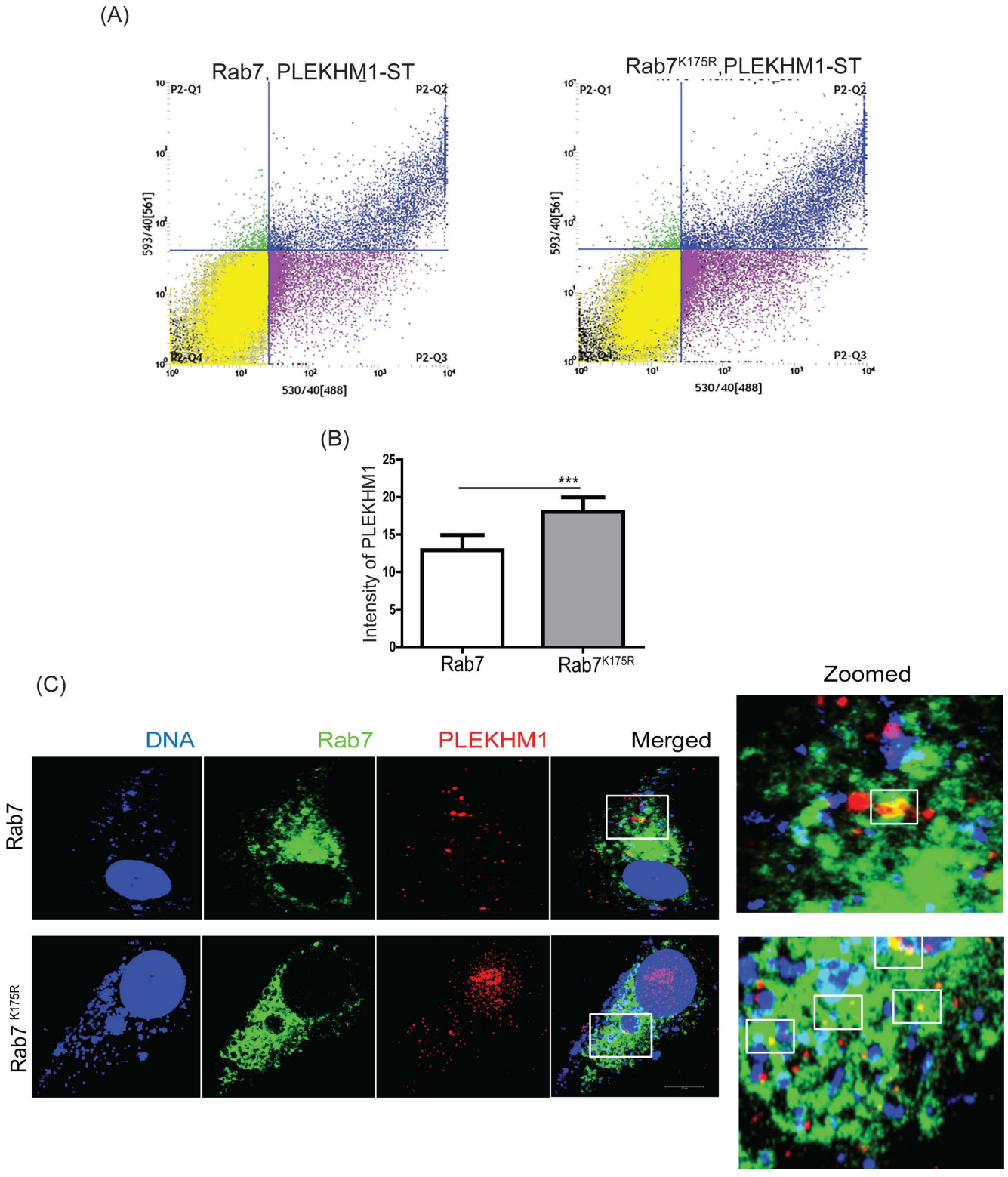


Fig. S4. Investigating the PLEKHM1 and Rab7 dynamics (A) HCT8 cells transfected with plasmids encoding WT-Rab7-GFP or K175R-Rab7-GFP along with Ds-Red PLEKHM1, were infected with ST for 4 hrs followed by Fluorescence Activated Cell Sorting (FACS) based analysis of various cell types having Rab7 (GFP) and/or PLEKHM1 (Ds-Red). (B) Graph represents the plots obtained for corresponding PLEKHM1 from three independent biological replicates. Mean±SEM from three experiments were plotted. The sign "***" indicates a P value of ≤ 0.001. (C) Confocal microscopic images showing the co-localization of Rab7 (Green) and Rab7^{K175R} (Green) with PLEKHM1 (Red) at 7 hr post infection. Bar-10μM.

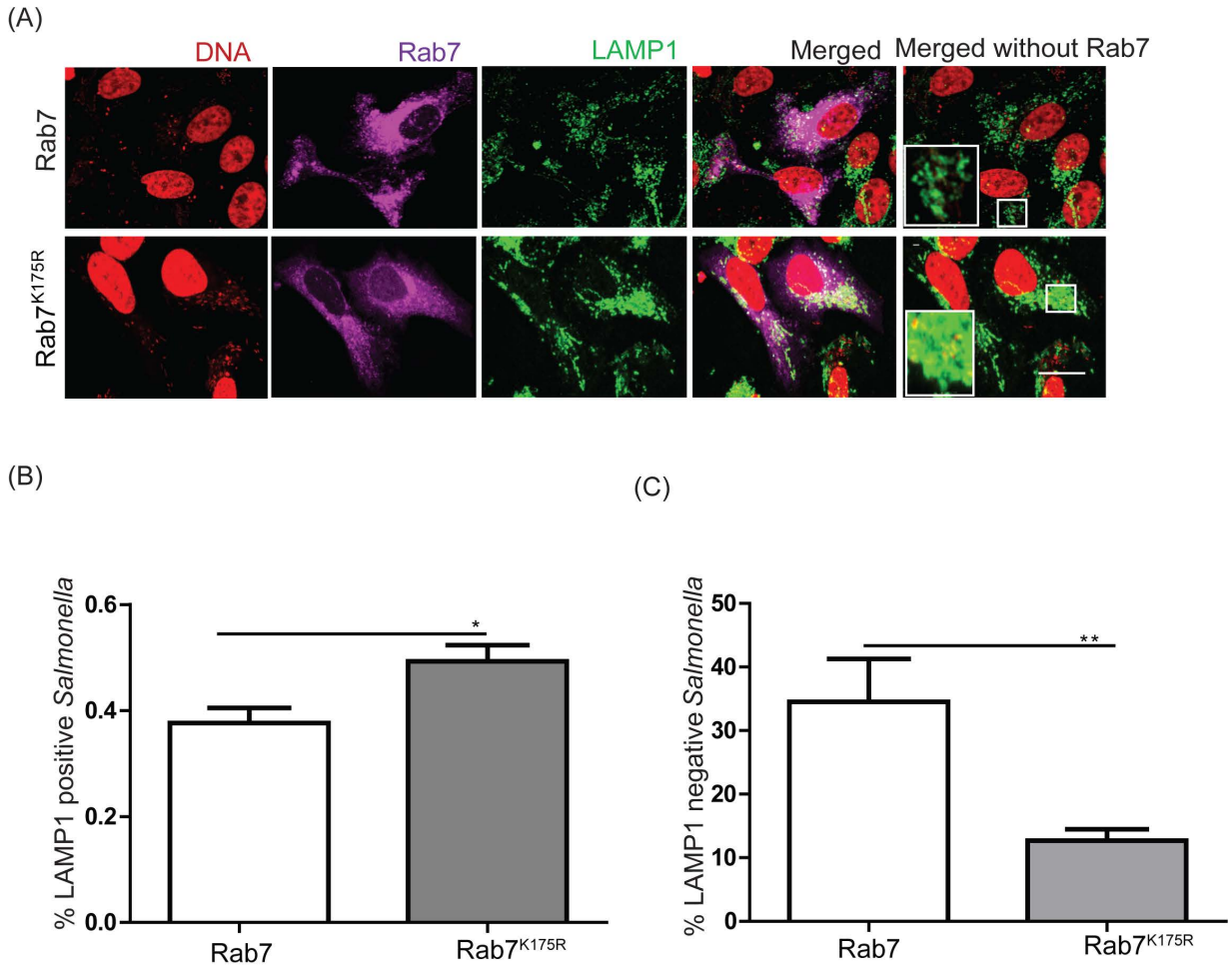
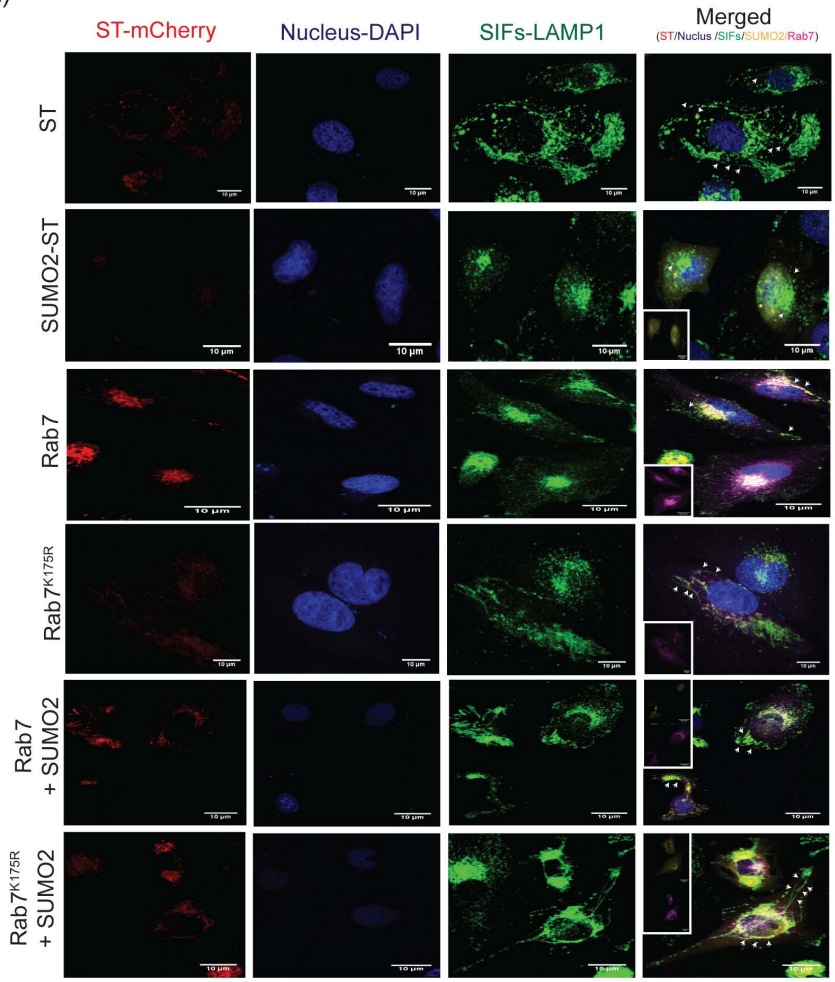
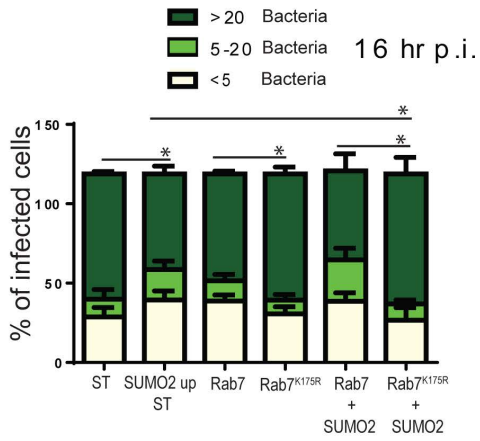


Fig. S5. SUMOylation of Rab7 modulates intracellular localization of ST (A) Confocal microscopic images of cells transfected with plasmids encoding Rab7 or Rab7^{K175R} that were infected with ST. Rab7 (magenta), LAMP1 for SCV (green) and DAPI for ST (Red) were imaged by confocal microscopy. Abundance of cytoplasmic ST and SCV resident ST was then calculated based on relative co-localization score of ST and LAMP1 (for SCV) versus free or dispersed ST. Areas of colocalization are highlighted by rectangular box within the image. Bar- 10µM. (B) Pearson's co-localization for the % of LAMP1 positive vesicles with ST as calculated using IMARIS software from three independent experiments. (C) Percent of ST (cytosolic) and its association with LAMP1 is calculated through visual observation from multiple confocal images denoted as % of LAMP1 negative ST. Mean±SEM from three experiments were plotted. Statistical analysis was carried out using Student's t test. The sign "*" indicates a P value of ≤ 0.05 and "**" indicates a P value of ≤ 0.01.

(A)



(B)



(C)

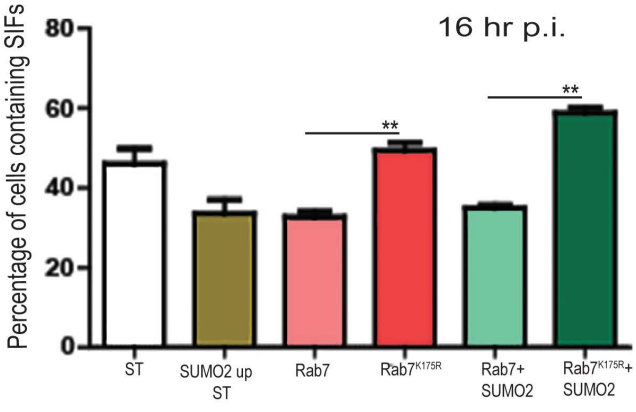


Fig. S6. Analysis of SIFs in cells with wild type and SUMO deficient Rab7. HeLa cells were transfected with plasmids encoding SUMO2/3 along with those encoding Rab7-WT or Rab7^{K175R} as indicated in the figure. These cells were infected with ST for 16 hrs and imaged for intracellular filament formation (SIFs) as shown in (A) Rab7-WT or Rab7^{K175R} seen in magenta, SUMO2 in yellow, ST in red and LAMP1 in green. (B) The number of intracellular bacteria was also scored in about ~50 individual cells. Number of bacteria were grouped as <5 bacteria/cell, 5-20 bacteria/cell or >20 bacteria/cell. The graph represents number of cells in each category plotted as mean percentage of total population using GraphPad prism software. Statistical analysis was carried in >20 bacteria/cell category. The sign "*" indicates a P value of ≤ 0.05 (C) The graph represents the number of SIFs obtained from confocal images. Mean ± SEM from three independent biological experiments were plotted. Statistical analysis was carried out by Unpaired Student's t test. "**" indicates a P value of ≤ 0.01.

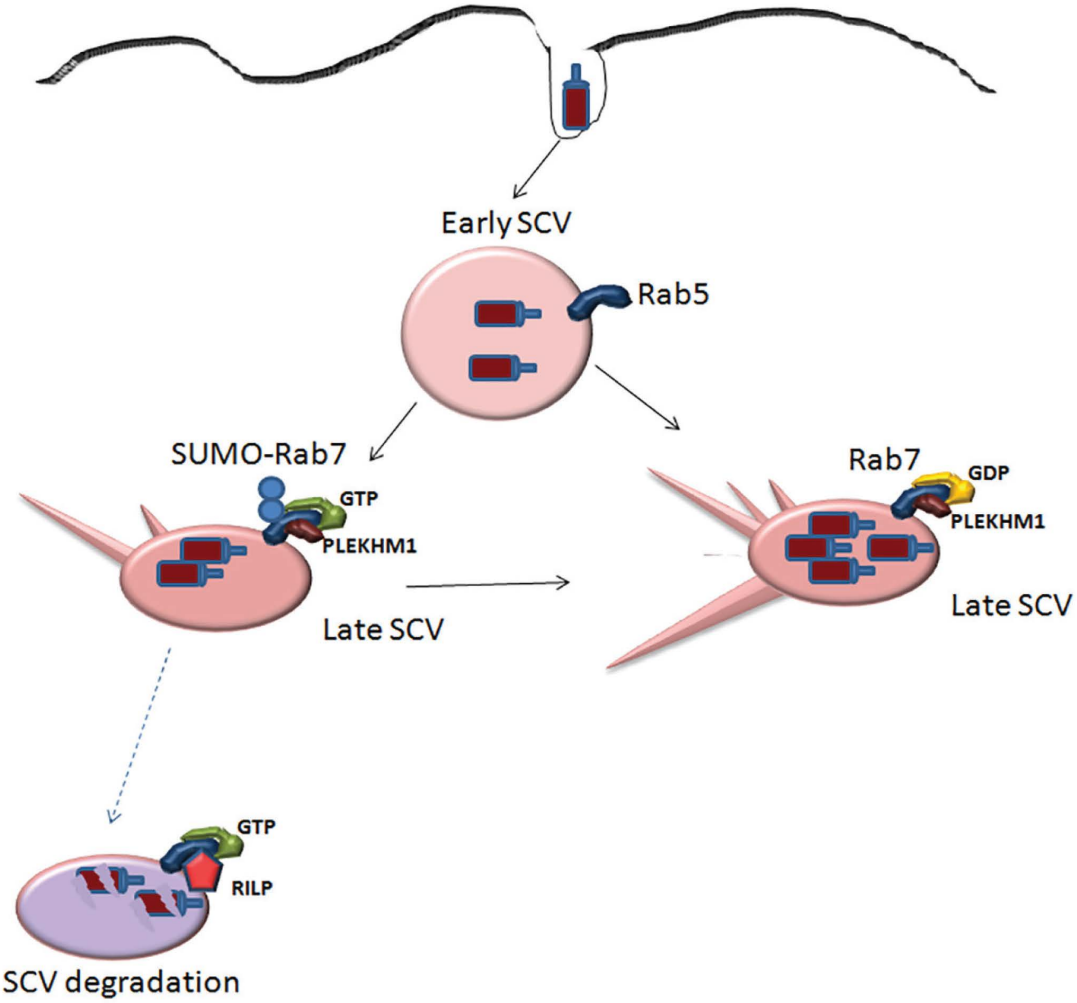


Fig. S7. Model depicting SUMOylation of Rab7 and connection to intracellular life of ST. Various components shown in the diagram may not be in scale.

Table S1. Number of Rab7 peptides detected from MS/MS from three biological replicates in uninfected control and *Salmonella* infected samples

	Uninfected Control			*Salmonella* infected samples		
Replicate →	I	II	III	I	II	III
	9	17	3	6	0	0

Table S2. Identification of site of SUMOylation of Rab7

Techniques employed to identify site of SUMOylation	**No. Of sites identified**	**Lysine positions**
High Score in silico analysis and MS/MS analysis	4	38,97,175, 194
Docking analysis	3	38,97,175
In vitro SUMOylation	1	175