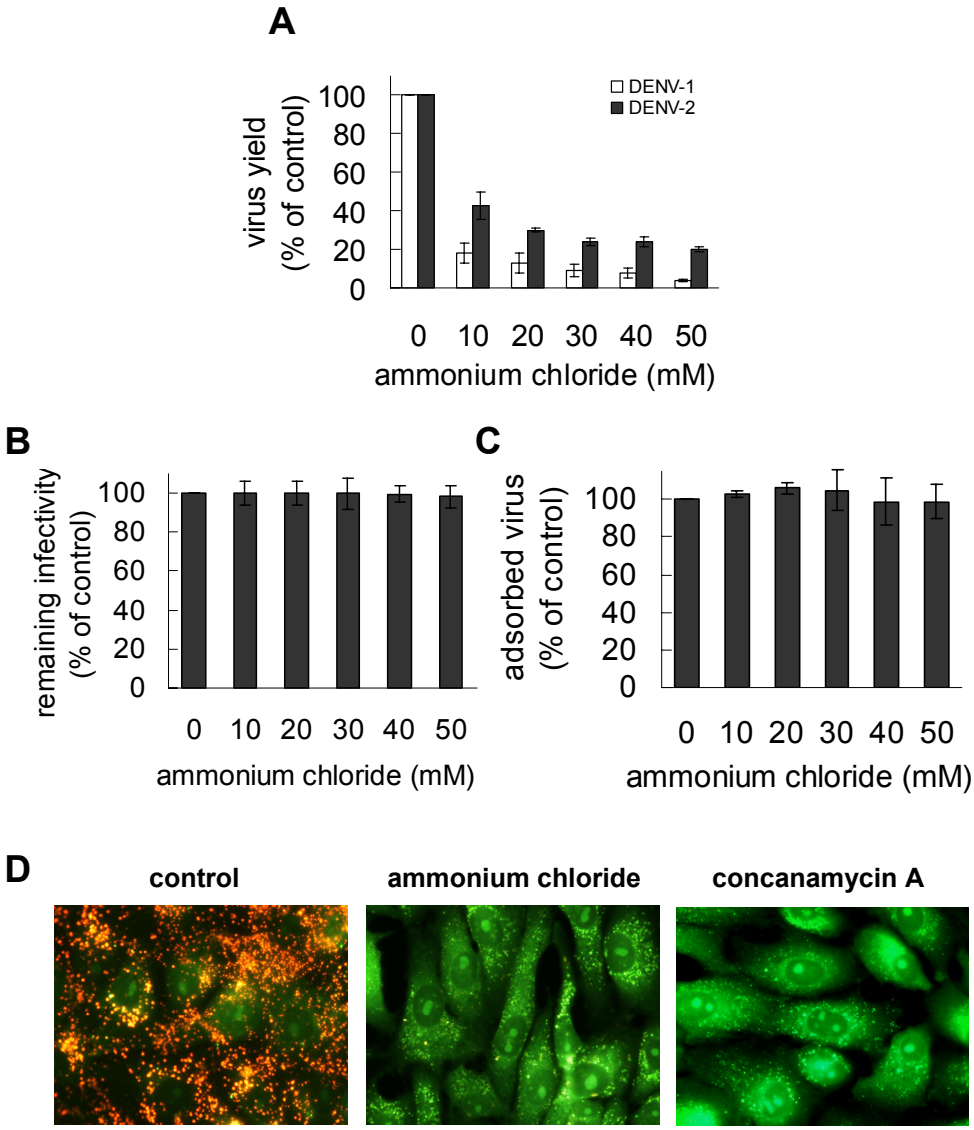

Fig. S1. Effect of ammonium chloride on DENV multiplication, viral particles infectivity, virus adsorption and intracellular vesicle acidification. A. Cells were treated with increasing concentrations of ammonium chloride and infected with DENV-1 or DENV-2. Virus yields were quantified by plaque assay at 48 h p.i. B. Suspensions of DENV-2 virions were incubated at 37 ºC with various concentrations of ammonium chloride. After 1 h samples were diluted and remaining infectivity was determined by plaque formation assay. C. Vero cells were treated as in A and infected with DENV-2 at 4 ºC. After 1 h adsorption in the presence or absence of the drug cells were disrupted and cell-bound virus was determined by plaque formation assay. In A, B and C Results are expressed as percentage of virus multiplication with respect to a control without drug treatment. Each point shows the mean of two independent experiments ± SD. D. Cells were left untreated (control) or treated with 50 mM ammonium chloride or 50 nM concanamycin A. Then, samples were stained with acridine orange.

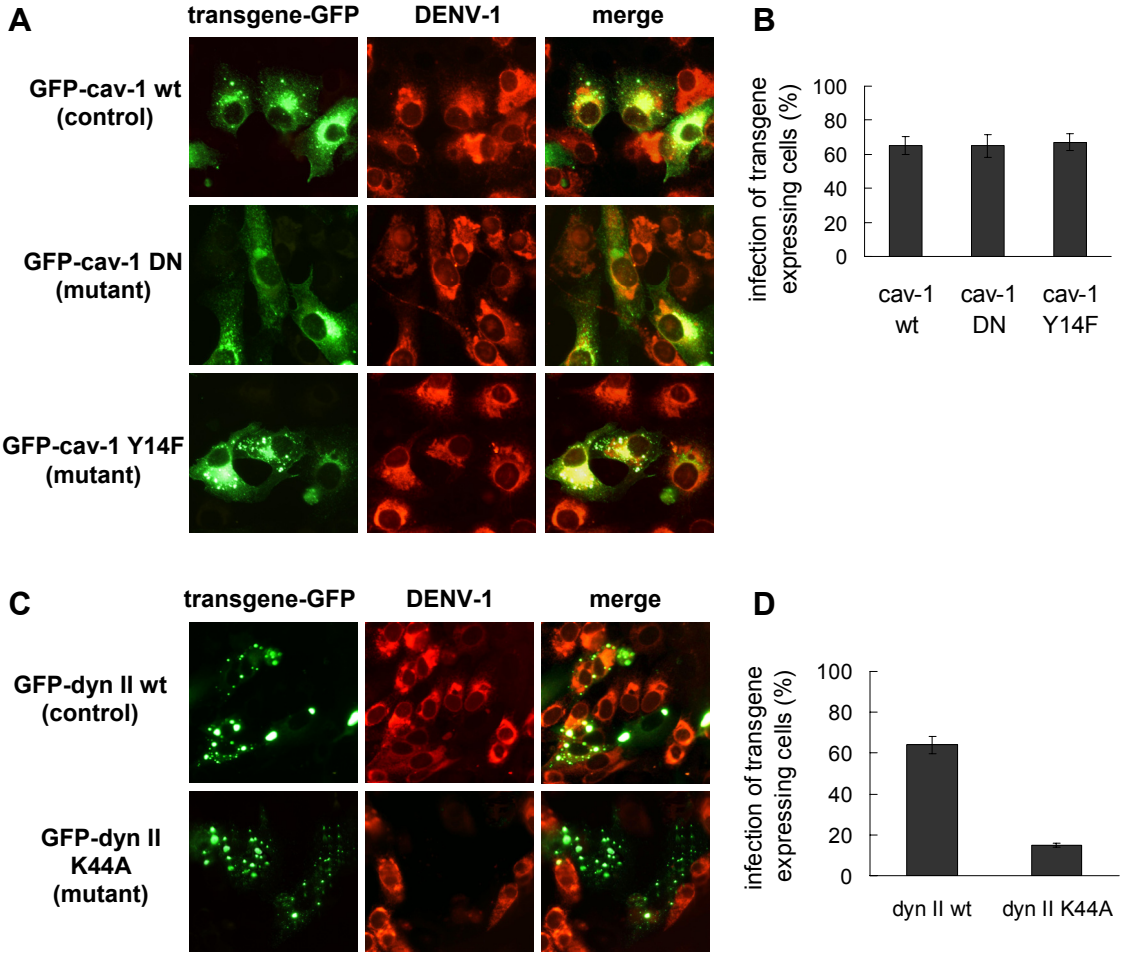

Fig. S2. DENV-1 infection of Vero cells is independent of caveolae but dependent on dynamin. A Cells transiently transfected with the constructs GFP-cav-1 wt, GFP cav-1 DN or GFP-cav-1 Y14F were infected with DENV-1. After 24 h infection cultures were fixed and immunofluorescence staining was performed. B. For quantification of samples shown in panel A, 250 transfected cells with similar levels of GFP expression were screened and cells positive for viral antigen were scored. C. Cells transiently transfected with the constructs GFP-dyn II wt or GFP-dyn II K44A were infected with DENV-1. After 24 h infection cultures were fixed and immunofluorescence staining was performed. D. For quantification of samples shown in panel C, 250 transfected cells with similar levels of GFP expression were screened and cells positive for viral antigen were scored. In B and D results are expressed as the mean of two independent experiments ± SD.

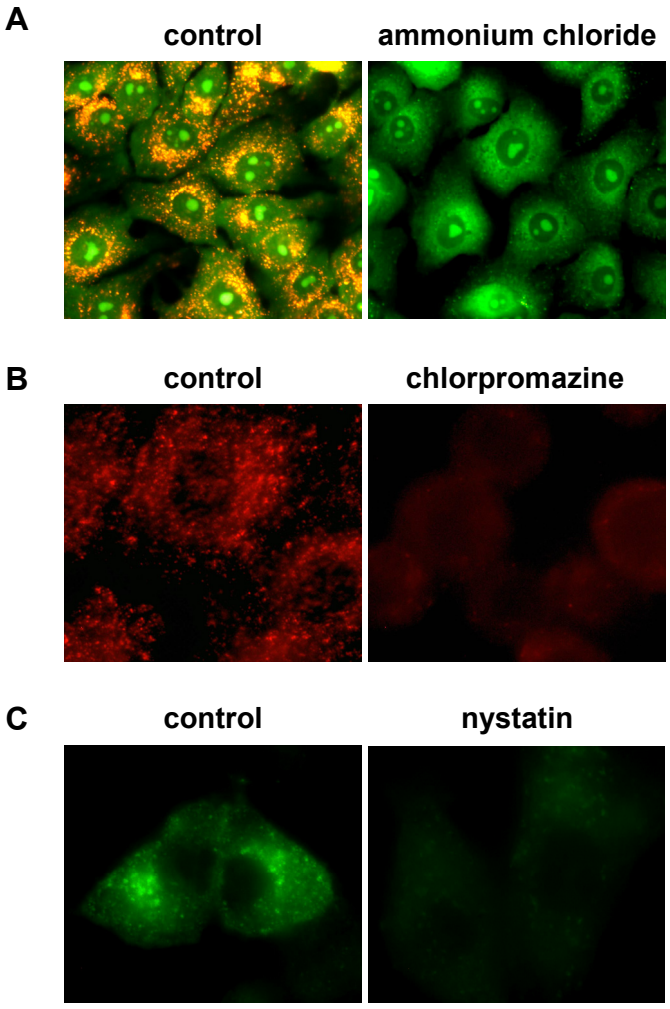

Fig. S3. Effect of different inhibitors on A549 cells endocytic processes. A549 cells were left untreated (control) or treated with ammonium chloride 50 mM (A), chlorpromazine 50 μM (B) or nystatin 100 μM (C). Then cells were stained with acridine orange (A), or incubated with TRITC-labelled transferrin (B) or FITC-labelled cholera toxin (C).