Supplemental Figures

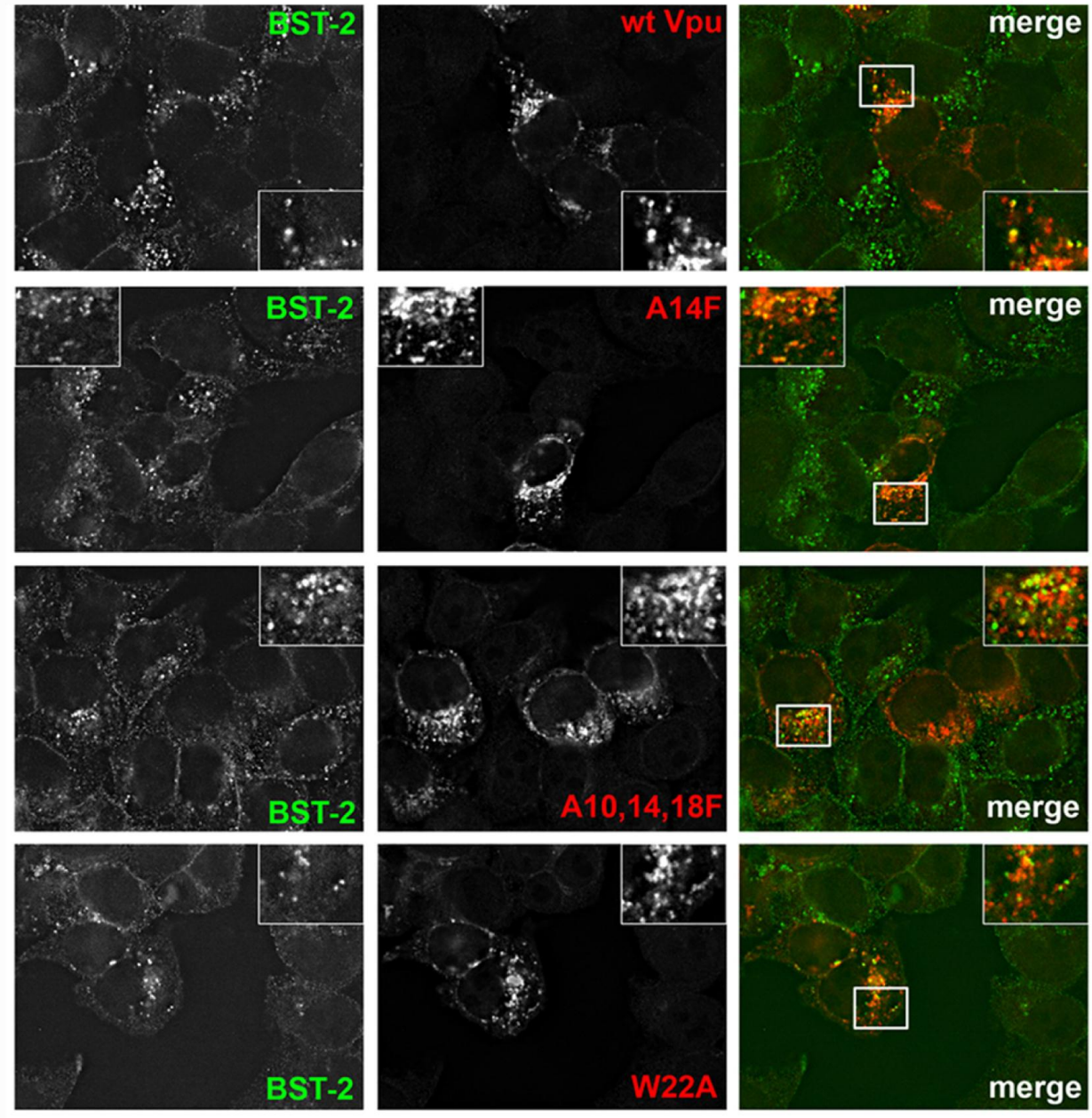

Figure S1. Vpu mutants of the AxxxAxxxAxxxW sequence colocalize normally with BST-2 in endosomes. HeLa cells, which express BST-2 constitutively, were transfected to express wild type Vpu or the indicated mutants. The next day the cell were stained using indirect immunofluorescence to detect Vpu (green) and BST-2 (red) as described previously(2).

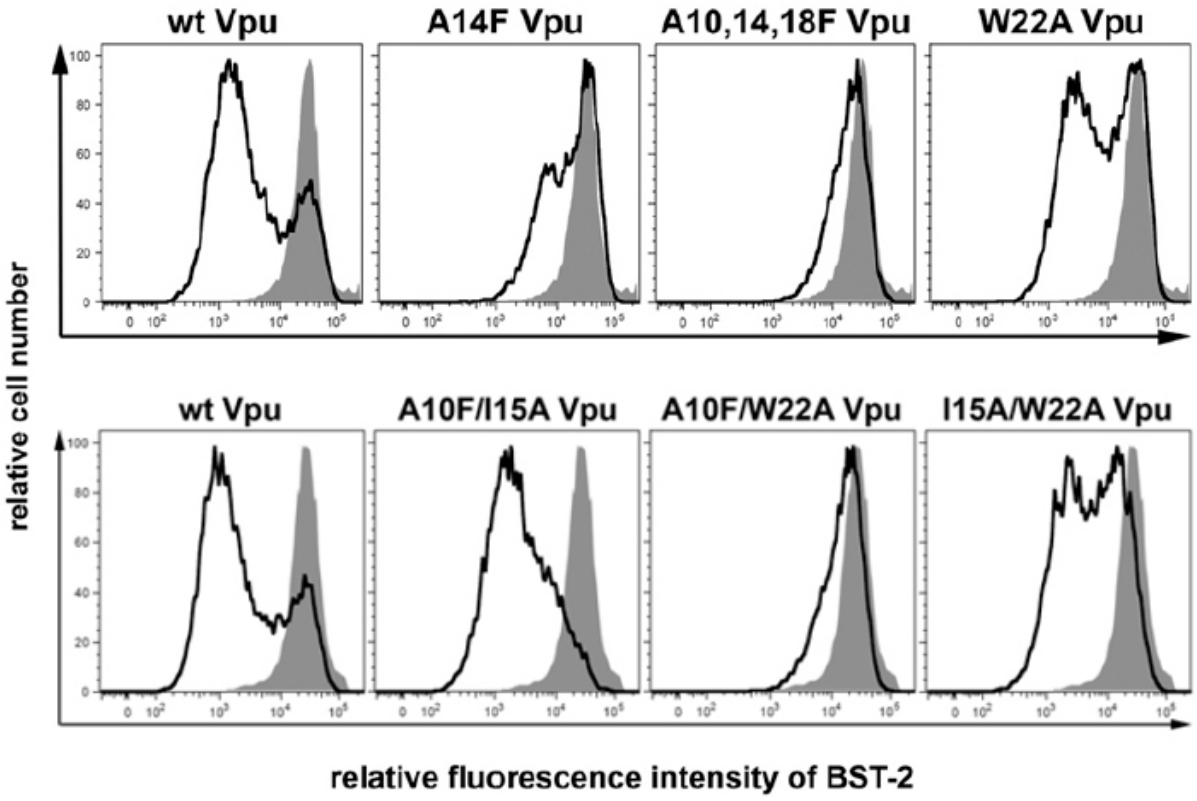

Figure S2. Top: Vpu-AAA/FFF, -A14F, and -W22A are impaired in their ability to down-regulate BST-2 from the cell surface. HeLa cells were transfected to express wild type Vpu or the indicated mutants along with GFP as a transfection marker. The next day the cells were stained for surface BST-2 using an indirect method and analyzed by two-color flow cytometry. Histograms are the relative cell number vs. BST-2 fluorescence intensity for the GFP-positive cells. Shaded curves are the empty vector control profile, which is identical in every panel. Open curves are derived from cells transfected to express wild type Vpu or the indicated mutants. Bottom: Vpu-A10F/W22A and to a lesser extent Vpu-I15A/W22A are impaired in their ability to downregulate BST-2 from the cell surface. The experiment was performed as described above.

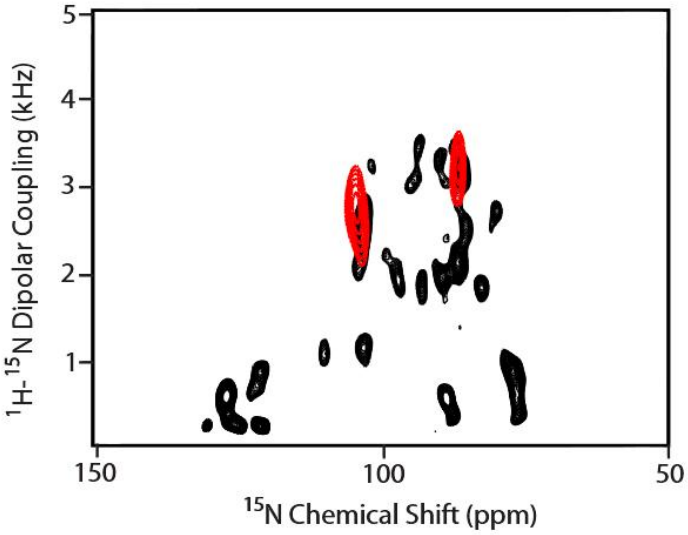

Figure S3. Overlay of ^{15}N chemical shift /^{1}H-^{15}N dipolar coupling separated local field spectra of uniformly ^{15}N labeled BST2-TMD(black) and ^{15}N valine labeled BST2-TMD (red).