Supplemental Figures:

Figure 1A. Cells cultured in Ca^{2+}-free medium were loaded with Fura-2AM and incubated with 1 μM thapsigargin followed by treatment with 5 μM A23187. Individual cells (n = 20) were imaged, and the average fluorescence intensity over time was recorded. The black and red arrows indicate the time point at which thapsigargin and A23187 were added respectively. Each experiment was repeated at least thrice.

Figure 1B. Cells cultured in Ca^{2+}-free medium were loaded with Fura-2AM and incubated with 1 μM thapsigargin followed by treatment with 500 μM glutamate. Individual cells (n = 24) were imaged, and the average fluorescence intensity over time was recorded. The black and red arrows indicate the time point at which thapsigargin and glutamate were added respectively. Each experiment was repeated at least thrice.

Figure 2. Quantification of the immunoblots of the EDTA eluates (left panel) and cell surface biotinylated extracts (right panel) from single mutants: **A**. Y23E. **B**. S25A. **C**. S25E.

For Y23F, no AnxA2-GFP band was detected and hence it couldn't be quantified.

Figure 3. Quantification of the immunoblots of the EDTA eluates (left panel) and cell surface biotinylated extracts (right panel) from double mutants: **A**. Y23ES25A and **B**. Y23ES25E

For Y23FS25A and Y23FS25E, no AnxA2-GFP band was detected and hence it couldn't be quantified.