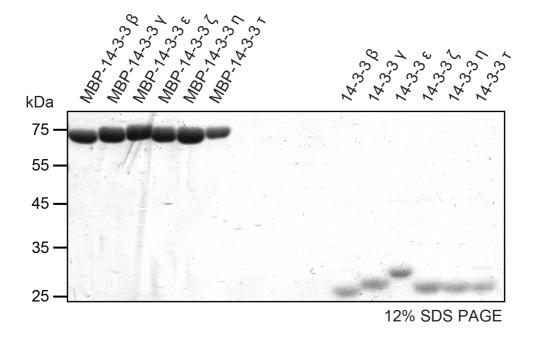
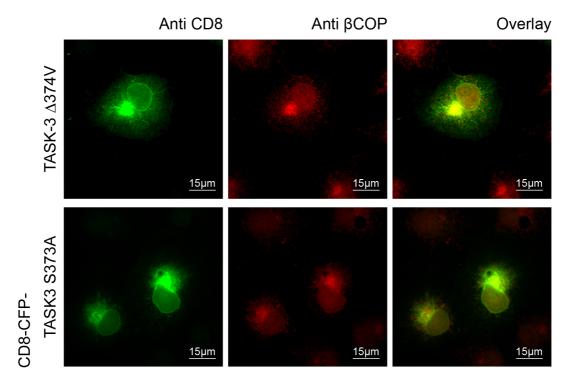
## **Supplemental Information**

## A dual phosphorylation switch controls 14-3-3-dependent cell surface expression of TASK-1

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**Fig. S1.** Coomassie-stained gel of six human 14-3-3 isoforms purified as MBP fusion proteins before (left) and after cleavage of the MBP tag and further purification by size exclusion chromatography (right). 2.5  $\mu$ g protein was loaded per lane.



**Fig. S2.** Subcellular localization of CD8-CFP-TASK-3  $\Delta$ V374 is indistinguishable from CD8-CFP-TASK-3 S373A. Constructs were transfected in three independent experiments and ca. 100 cells per transfection were analyzed by indirect immunostaining against the indicated antigens.

**Table S1.** Binding affinities of 14-3-3 proteins to TASK-1 C-terminal peptides determined by FP. Equilibrium dissociation constants  $(K_D)$  are displayed in [nM]. Error is s.e.m.

FP  $K_D [\mu M]$ TASK-1 14-3-3 **TASK-1 pS392 TASK-1 pS393** isoform pS392pS393 7.5 0.7 β 0.1 33.1  $92.2 \pm$ 2.9 9.8  $\pm$ 0.6 51.4  $\pm$ 2.7  $190.4 \pm$ 19.0 γ 29.0 373.6 1.0 33.2 861.4 23.0 3 23.0 1.4 133.0 1.8  $264.4 \pm$ 8.9  $9.2 \pm$ 0.1 81.5 7.8  $525.4 \pm$ 120.0  $\pm$ η 2.0 9.0 49.4  $\pm$ 123.2  $662.9 \pm$ 86.3 τ  $36.0 \pm$ 2.9  $58.8 \pm$ 5.3 106.2 ± 10.4 σ

Table S2. Antibodies used in this study.

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Table S3. Plasmids used in this study.

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Table S4. Primers used in this study.

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