

**Figure S1** The direct Pho85 target , Pho4, is desphosphorylated upon Pho85 inhibition with 1NM-PP1 and shuttled from the cytoplasm to the nucleus. *pho85-as/cdc28as* strain was transformed with an expression plasmid containing a Pho4p-GFP fusion construct under the control of the ADH1 promoter (BA1828, B. Andrews lab). Cells were grown to an OD<sub>600</sub> of 0.5 (0h) and treated with DMSO or 25  $\mu$ M 1NM-PP1. Cellular localization of Pho4p was analyzed using fluorescence microscopy at 15 min after treatment. Representative fields are shown.



**Figure S2** Abp1p expression is not cell cycle regulated. (A) Flow cytometry analysis of DNA content of wild type yeast cells synchronized by release from  $\alpha$ -factor at the indicated times in the left. (B) Protein extracts were analyzed by Western blotting using polyclonal anti-Abp1p antibody to monitor Abp1p levels or anti-coronin antibody to control for cellular protein levels. Time is minutes after  $\alpha$ -factor release. The positions of phosphorylated and unphosphorylated Abp1p are indicated.





**Figure S3** Representative Western blots of Abp1p expression in strains bearing single deletions of non-essential kinases (A) or over-expression of essential kinases (B). (A) Western blot analysis, using polyclonal anti-Abp1p antibody, of yeast extracts from kinase deleted strains backgrounds. (B) Western blot analysis, using polyclonal anti-Abp1p antibody, of yeast extracts from transformants expressing various essential kinases from the *GAL1* promoter. Extracts were made from mid-log phase cells grown in presence of glusose (D) or galactose (G).