

Figure S3. Characterization of the non-phosphorylable and phospho-mimetic mutants of Cdc55. (a) Cdc55 phospho-mutants rescue the cold sensitivity of $cdc55\Delta$ strain. Strains Y986, Y1281, Y1284 and Y1294 were grown at 25°C and 14°C for 3 days. (b) Cdc55 phospho -mutants rescue the elongated morphology of $cdc55\Delta$ strain. Cycling cells of strains as in (a) were grown at 25°C. Representative images are shown. (c) Cdc55 phospho-mutants physically interact with the catalytic subunit Pph21. Cells extracts were prepared from strain Y616 containing the pRS316 (empty vector), E427 (*Pk3-CDC55*), E430 (*Pk3-CDC55-2A*) and E463 (*Pk3-CDC55-ED*). Co-immunoprecipitation of Cdc55 with Pph21 was analyzed. A representative experiment of one of the assays is shown (left). A quantification of three independent experiments is depicted (right). Means and SD are shown. (d) Cdc28-Y19 phosphorylation is not altered in the Cdc55 phospho-mutants. Cell extracts from strains as in (a) in cycling cells were prepared. Phosphorylation of Cdc28-Y19 and Cdc28 protein levels were detected by western blot. A representative experiment is shown (left). A quantification of three independent cultures, means and SD are shown (right).