



Nishiyama *et al.* Supplementary Figure 4

Supplementary Figure 4

Determination of the xTRF1 region phosphorylated by Plx1 *in vitro*

(A) Structures of xTRF1 mutant proteins used for the *in vitro* kinase assay with Plx1. The TRFH (residues 9-209) and Myb (residues 367-420) domains are indicated. The results of the *in vitro* kinase assay for each mutant recombinant protein, which are shown in B and C, are summarized.

(B) An NH₂-terminal region (residues 1-205) and a COOH-terminal region (residues 206-420) fused with histidine hexamer at the N-terminus, were expressed in and purified from *E. coli* and tested for phosphorylation by immunoprecipitated Plx1 *in vitro*. CBB staining (left) and autoradiography (right) are shown.

(C) A series of GST-tagged xTRF1 fragments were expressed in *E. coli* and purified with glutathione-Sepharose 4B beads. These proteins were examined by phosphorylation assay with immunoprecipitated Plx1 *in vitro*. CBB staining (bottom panel) and autoradiography (top panel) are shown.