





Figure S1. The RTA variants preG212E and preP95L/E145K are not substrates for Png1. (A) The viability of BY4743 and  $png1\Delta$  expressing preG212E and preP95L/E145K. A series of ten-fold dilutions were spotted on glucose and galactose plates after overnight growth in glucose. (B) Immunoblot analysis of BY4743 and  $png1\Delta$  expressing preG212E and preP95L/E145K. The membrane fraction (M) and cytosol fraction (C) isolated at 6 and 24 hpi were separated on a 10% SDS-polyacrylamide gel and probed with monoclonal anti-RTA (1:5000). The ER membrane marker Dpm1p and cytosolic marker Pgk1p were used as loading controls. (C) Ribosome depurination by wild type preRTA, preG212E and preP95L/E145K expressed in BY4743 and  $png1\Delta$  by qRT-PCR at 4 hpi.