

**Figure S2** An H2B K123R mutant strain is sensitive to cycloheximide but does not exhibit increased expression of a nonstop reporter. (A) HTB1 (KY2043) and htb1-K123R (KY2044) strains were grown to saturation and diluted to  $3 \times 10^8$  cells/ml. Ten-fold serial dilutions were performed on YPD or YPD containing 0.8 µg/ml cycloheximide (CHX) and incubated at  $30^{\circ}$ C for 2 or 6 days, respectively. (B) HTB1 (KY2043) and htb1-K123R (KY2044) strains were transformed with a LEU2-marked plasmid containing a his3 nonstop reporter. Strains were then grown to saturation and diluted to  $1 \times 10^8$  cells/ml. Ten-fold serial dilutions were performed on SC-L or SC-L-H and plates were incubated at  $30^{\circ}$ C for 3 days.