

B


| Sld3 ${ }^{\text {Esal }}$ (-)-1 | Y178F'N189Y | S459L |
| :---: | :---: | :---: |
| SId3 ${ }^{\text {Esal }}$ (-)-2 | R192L |  |
| SId3Esal(-)-3 | P1574 $T^{19}$ |  |

P623Q

## C

AD-Esa1


| Clone \# | Viable? | Name |
| :---: | :---: | :---: |
| \#15 | No |  |
| \#32 | Yes | -> Esa1 ${ }^{\text {Sld3 (-)-1 }}$ |
| \#37 | Yes | -> Esa1 ${ }^{\text {Sld } 3(-)-2}$ |



## Supplementary Figure S4. Isolation of mutants that diminish the SId3-Esa1 interaction.

A. Interaction between SId3 and various Esa1 constructs, Eaf5, and Eaf6 were tested in the Y2H analysis. AD: activation domain. DB: DNA-binding domain. B. Schematics of SId3Esa1(-) mutant proteins. Numbers indicate the positions of amino acid residues. C. Isolation of Esa1SId3(-) mutants. Interactions between Esa1SId3(-) candidates and SId3 were tested in the Y2H. Clone \# indicates the initial number of candidates. c: Positive control (Esa1) for the Y2H. Clone \#15 lacked an interaction with SId3 in this assay, as did \#32 and \#37. However, plasmid shuffling assay revealed that \#15 could not support cell growth, whereas \#32 and \#37 could. Therefore, \#15 was not retained as an Esa1SId3(-) mutant and \#32 and \#37 were named Esa1 ${ }^{\text {Sld }(-)-1}$ and -2 , respectively. (see Materials and Methods). D. Schematics of Esa1 ${ }^{\text {Sld }}$ (-) mutant proteins Numbers indicate the positions of amino acid residues.

