

Supplemental figure 2: Complementation assay using a *GAL::rrp44* strain confirms that the N-terminal region of Rrp44p is important for exosome function.

To confirm the complementation results seen in the *rrp44* Δ strain, the truncations described in the main text were tested in a strain containing *RRP44* under the control of the *GAL10* promoter (*GAL::rrp44*)¹. In this strain the *RRP44* gene is actively transcribed when the strain is grown on galactose as the sole carbon source. However, growth on glucose represses the *GAL10* promoter, and thus expression of *RRP44*. It has previously been reported that this *GAL::rrp44* strain fails to grow on glucose¹, but we observed slow growth, suggesting that low levels of Rrp44p remain. Each of the *RRP44* truncations was introduced into this strain. Similar to what was seen in the *rrp44* Δ strain, both full length and the S1 domain deletion restored close-to-wild type growth rates to the *GAL::rrp44* strain. Surprisingly, however, several versions of Rrp44p that are missing the RNB domain that supported very slow growth in the *rrp44* Δ strain supported much faster growth of the *GAL::rrp44* strain. As expected, further C-terminal truncation, or any truncation from the N-terminus failed to support growth. These results confirm that the previously uncharacterized N-terminal region is an important part of the exosome. Furthermore, these results suggest that Rrp44p may have two distinct functions. One function requires the N-terminus and is essential. The second function is important for normal growth rates and requires either the RNase II homologous region, or very low levels of the full length protein.

1. Mitchell, P., Petfalski, E., Shevchenko, A., Mann, M. & Tollervey, D. The exosome: a conserved eukaryotic RNA processing complex containing multiple 3'→5' exoribonucleases. *Cell* **91**, 457-66. (1997).

GAL::rrp44 +

Glucose

Galactose



Vector

Vector

