Zhang et al. http://www.jcb.org/cgi/doi/10.1083/jcb.200502055

Methods for the rescuing experiment

To test whether overexpression of Sro7p rescues the deletion of *EXO84* and *EXO70*, two strains were generated: GY1264 and GY2180 (Table I). In GY1264, the endogenous *EXO84* gene was disrupted by *HIS3* and was supplemented with a *CEN URA3 EXO84* plasmid as a balancer. Likewise, in GY2180, the endogenous *EXO70* was replaced by *HIS3* and was supplemented with a *CEN URA3 EXO70*. For *SRO7* overexpression, a *2μ LEU2 SRO7* plasmid was transformed into GY1264 and GY2180. As positive controls, *CEN* plasmids containing *LEU2 EXO84* or *LEU2 EXO70* were also transformed to GY1264 and GY2180, respectively. The *2μ LEU2* vector was transformed into the two host strains as a negative control. SC Leu plates containing uracil and 5-FOA (1 mg/ml at final concentration) were used to test whether the cells, with high-copy *SRO7*, were able to survive after losing the *CEN URA3* balancer plasmid carrying *EXO84* or *EXO70*.