Balanced trafficking between the ER and the Golgi apparatus increases protein secretion in yeast - DTU Orbit (13/11/2018)

**Balanced trafficking between the ER and the Golgi apparatus increases protein secretion in yeast**

The yeast *Saccharomyces cerevisiae* is widely used as a cell factory to produce recombinant proteins. However, *S. cerevisiae* naturally secretes only a few proteins, such as invertase and the mating alpha factor, and its secretory capacity is limited. It has been reported that engineering protein anterograde trafficking from the endoplasmic reticulum to the Golgi apparatus by the moderate overexpression of SEC16 could increase recombinant protein secretion in *S. cerevisiae*. In this study, the retrograde trafficking in a strain with moderate overexpression of SEC16 was engineered by overexpression of ADP-ribosylation factor GTP activating proteins, Gcs1p and Glo3p, which are involved in the process of COPI-coated vesicle formation. Engineering the retrograde trafficking increased the secretion of alpha-amylase but did not induce production of reactive oxygen species. An expanded ER membrane was detected in both the GCS1 and GLO3 overexpression strains. Physiological characterizations during batch fermentation showed that GLO3 overexpression had better effect on recombinant protein secretion than GCS1 overexpression. Additionally, the GLO3 overexpression strain had higher secretion of two other recombinant proteins, endoglucanase I from *Trichoderma reesei* and glucan-1,4-alpha-glucosidase from *Rhizopus oryzae*, indicating overexpression of GLO3 in a SEC16 moderate overexpression strain might be a general strategy for improving production of secreted proteins by yeast.

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