

**Background/objective**

Glucose and xylose are the two most abundant sugars in lignocellulosic biomass, and simultaneous co-fermentation of glucose and xylose is a key desired trait of engineered *Saccharomyces cerevisiae* for efficient and rapid production of biofuels and chemicals. However, the presence of glucose strongly inhibits xylose transport in *S. cerevisiae*'s endogenous hexose transporters. Here we used bioprospecting in previously unexplored oleaginous yeasts and plants to identify sugar transporters *Lipomyces starkeyi* LST1\_205437 and *Arabidopsis thaliana* AtSWEET7 capable of co-transporting glucose and xylose in yeasts.

**Approach**

- ❖ Used engineered *S. cerevisiae*, that were capable of xylose fermentation but lacked major glucose transporters, to screen for and characterize transporters capable of co-fermenting both sugars.
- ❖ Performed kinetic studies using <sup>14</sup>C labeling to understand the kinetic background behind simultaneous glucose and xylose co-fermentation.
- ❖ *In silico* molecular modeling and dynamics simulations (MD) were used to explain the molecular basis of this unique trait observed in the selected transporters.

**Results**

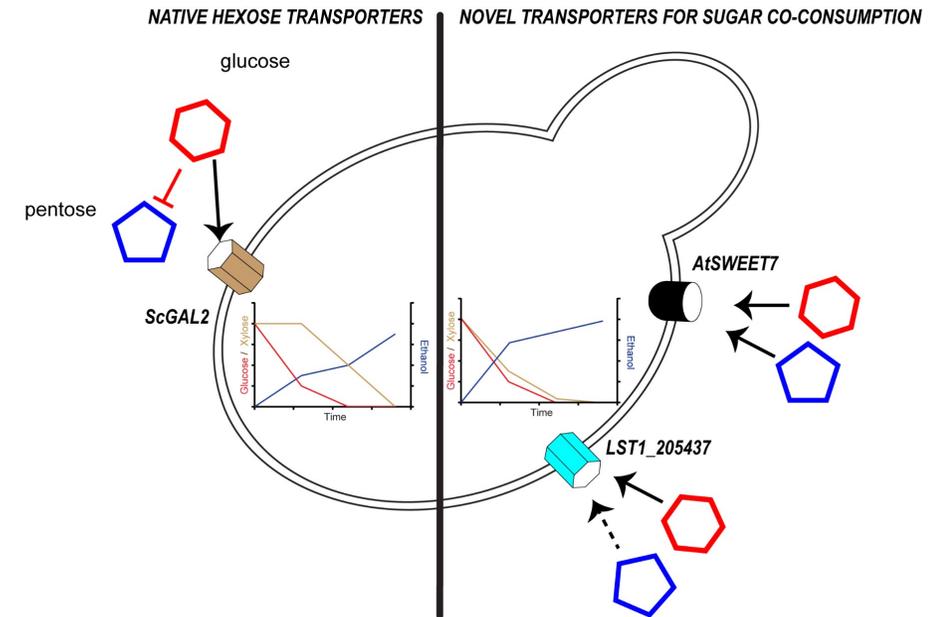
- ❖ Identified two transporters, *L. starkeyi* LST1\_205437 and *A. thaliana* SWEET7, with an ability to transport glucose and xylose simultaneously.
- ❖ Kinetics studies revealed that both transporters transport xylose in the presence of glucose.
- ❖ MD modeling studies revealed the involvement of several key amino acids at sugar binding sites that can accommodate both glucose and xylose.

**Significance**

These findings provide new insight on the glucose and xylose transport mechanism of sugar transporters, and the identified sugar transporters can be employed to develop engineered yeast strains for producing cellulosic biofuels and chemicals.

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# Identification and Analysis of Sugar Transporters Capable of Co-Transporting Glucose and Xylose Simultaneously



**Ability of two structurally distant transporters with 7 and 12 transmembrane (TM) domains to facilitate partial and complete co-transportation of glucose and xylose in *S. cerevisiae*.**