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Expression of a Bacterial Trehalose 6-phosphate Synthase Gene *otsA* in *Camelina sativa* Seeds Promotes the Channeling of Carbon Towards Oil Accumulation

Background/Objective

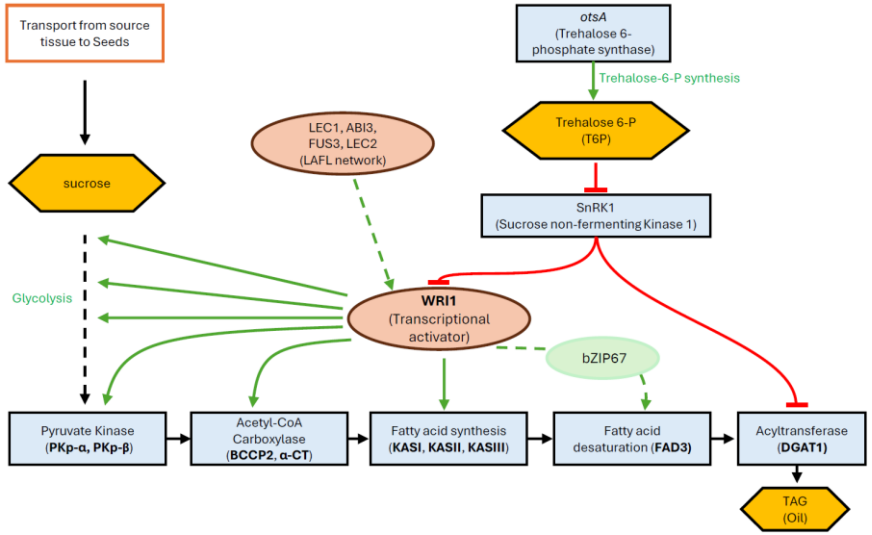
Improving seed oil yield is essential for developing *Camelina sativa* as a sustainable biofuel crop. Fatty acid synthesis depends on the production of acetyl-CoA, from photosynthetically derived sugars. Trehalose 6-phosphate (T6P), a proxy for sucrose availability, can link sugar status to plant growth and development. Our previous studies on Arabidopsis transgenic lines constitutively expressing the *E. coli otsA* (encoding TPS) showed increased T6P levels and seed triacylglycerol, along with stunted growth. In this study, we examined the effects of selectively elevating T6P in seeds of the emerging oilseed crop.

Approach

We expressed *otsA* in camelina under the control of a seed-specific Phaseolin promoter. We manipulated sugar signaling by the seed-specific expression of *E. coli otsA* to elevate T6P levels in developing seeds of camelina. We hypothesized that expressing the bacterial *otsA* in seeds would enhance fatty acid synthesis and TAG accumulation without eliciting the detrimental pleiotropic effects associated with its constitutive expression.

Results

Seeds of the resulting transgenics lines accumulated high levels of T6P, and a 15–20% increase in total fatty acids and triacylglycerol compared to wild-type. Molecular analysis showed the transgenic seeds had reduced SnRK1 activity, elevated WRI1 protein levels, and increased transcript levels of its target genes along with enhanced rates of fatty acid synthesis that increased seed weights relative to wild type. Crucially, seed-specific expression of *otsA* mitigated the growth defects associated with constitutive otsA expression, and the transgenic lines showed normal seed germination and establishment.



Proposed model of Trehalose-6-Phosphate (T6P) - mediated enhancement of oil biosynthesis

Significance/Impacts

These findings demonstrate that targeted T6P modulation via seed-specific *otsA* expression is an effective metabolic engineering strategy to boost oil production in camelina and potentially in other oilseed crops and bioenergy crops such as energycane, sorghum, and miscanthus. Further work is needed to investigate whether elevating seed T6P levels influences other aspects of physiology in response to biotic and abiotic stresses.

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