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Improving the thermal stability of Rubisco activase

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Summary

The major objective is to produce wheat germplasm with improved tolerance to heat stress through the modification of the key photosynthetic enzyme Rubisco activase. Current funds have allowed transformation of “best bet” candidate gene constructs into wheat as part of SP1.5. Joanna Scales, a BBSRC PhD student jointly supervised by Martin Parry, Christine Raines, and Mike Salvucci, generated the transformation constructs and will undertake the molecular and biochemical analysis of transgenic lines.

Results and Discussion

Higher temperatures (a likely result of climate change) induce inactivation of Rubisco, compromising the stability of yield improvements achieved through increases in photosynthetic capacity and efficiency in projects SP1.1, SP1.2, and SP1.7. The heat stability of Rubisco activase, the protein regulating Rubisco activity in wheat leaves, is an important determinant of the heat stability of photosynthesis (Salvucci and Crafts-Brandner 2004). This project will test the hypothesis that transgenic wheat containing an engineered Rubisco activase, previously shown to be heat stable *in vitro*, will possess this trait in *planta* under field conditions. This project is high impact with a medium- to long-term delivery timeframe.

USDA experiments have confirmed that Rubisco activase from cotton has a high thermal tolerance. Recent experiments have confirmed that, *in vitro*, purified wheat Rubisco (supplied by Rothamsted Research) is successfully activated by recombinant Rubisco activase from cotton. Thus, the duo cotton Rubisco activase and wheat Rubisco is expected to function *in vivo*.

We have designed constructs to introduce the thermally stable cotton Rubisco activase into wheat (Figure 1). This has been used in transformation

experiments and putative transgenic lines are under selection. Lines expressing the transgene will be tested for photosynthetic function at high temperature.

Next Steps

We will analyse the transgenic lines and confirm that the thermally stable Rubisco activase is expressed, correctly imported, and functional in the mesophyll chloroplasts. If the expression of cotton Rubisco activase is not confirmed, we will redesign the construct to alter the promoter and/or transit peptides, and generate a new construct and transgenic lines. Once transgenic lines successfully expressing the thermally stable Rubisco activase at different temperatures are achieved, we will analyse their photosynthetic performance and test lines with improved thermal tolerance in controlled environment then field conditions.

Reference

Salvucci, M.E. and S.J. Crafts-Brandner. 2004. *Physiologia Plantarum* 120:179–186.

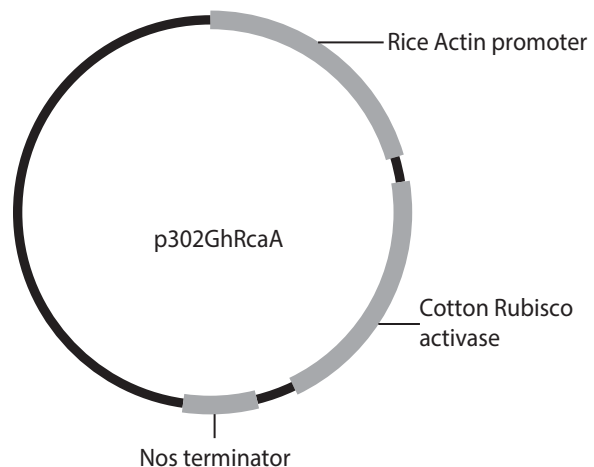


Figure 1. Plasmid p302GhRcaA for expression of cotton Rubisco activase in wheat.