Directed evolution of Rubisco for improved carbon dioxide capture and conversion Tina Wang (PI) Department of Chemistry University of Wisconsin–Madison		 Research objectives Improve plant Rubisco through directed evolution to achieve 5-fold increase in k_{cat}^C while maintaining CO₂ specificity SOA: Modest (< 2-fold) improvements in bacterial or algal Rubiscos using flawed methodology Innovation: New platform for Rubisco directed evolution based on direct detection of reactivity, 	
		 Higher carboxylation I Maintain CO₂ specific Compatible with plant biogenesis 	address Rubisco biogenesis during evolution, evolve plant Rubiscos • Start: TRL1 • End: TRL2 exit criteria met
 Approach Develop new selection platform that monitors Rubisco reactivity with RuBP and 2PG biosensors 	RuBP	O_2 $3PGA + 2PC$	 Potential impact Provide a platform for directed evolution of Rubiscos from variety of organisms
 Increase Rubisco folding and assembly during directed evolution through chaperonin/assembly factor co-expression Engineer assembly factors to tolerate mutations in Rubisco large subunit substrates Evolve <i>Arabidopsis thaliana</i> Rubisco (AtRubisco) using new selection and improved methodology After activity evolution, re-optimize for compatibility with native AtRubisco biogenesis machinery 		 Improve photosynthetic efficiency in plants Reduce power and water use requirements by increasing Rubisco carboxylation rates Reduce Rubisco leaf protein investment to improve harvest index Increase crop yields for both for space exploration and agriculture on Earth 	