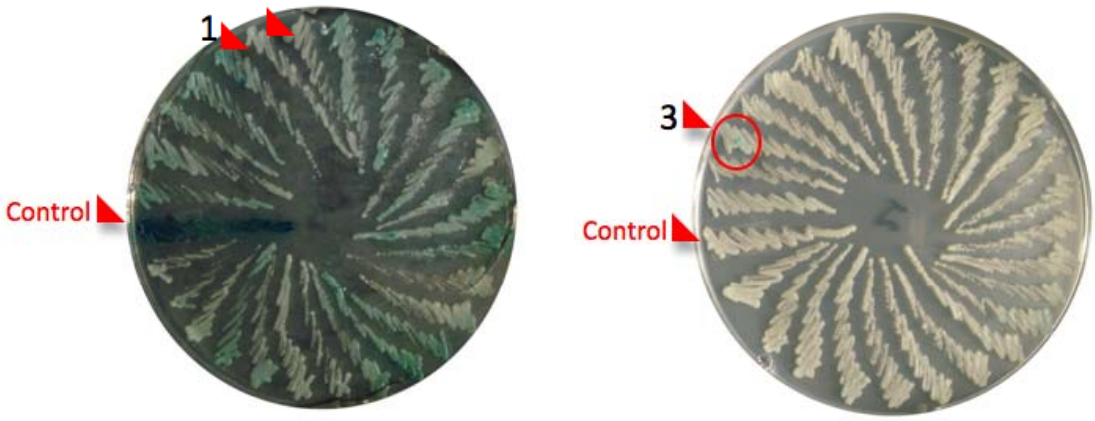
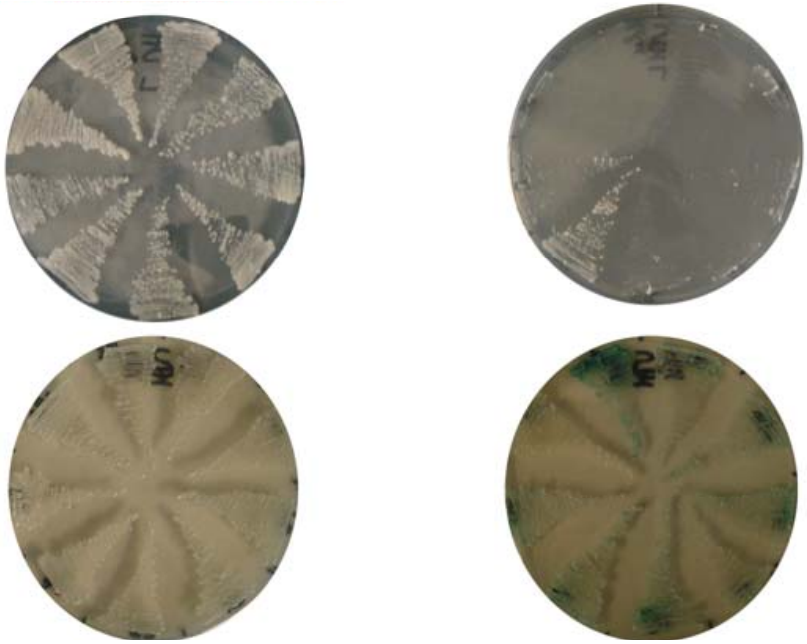
(A)



(B)



(C)

Figure S1. Procedure of Y2H screening for proteins interacting with the AtCERK1

(A) Testing for interaction with the galactose-dependent *lacZ* reporter gene

Colonies growing on selection medium Gal(UTHL)$^-$were streaked onto selection medium Gal-X gal-(UTH)$^-$(left panel) and Glc-X gal (UTH)$^-$(right panel) to test for the ability to activate galactose-dependent LacZ expression. Images were acquired one week after colony streaking. Eg. (1) and (2) did not turn blue on the Gal-X gal-(UTH)$^-$medium, indicating that there is no protein interacting with the kinase domain of CERK1 (K1) to activate LacZ expression. In addition, (3) is a false positive clone because the colony turned blue on X gal medium regardless of carbon source (the *lacZ* gene is under a Gal promoter).

(B) Testing of individual interactors with CERK1

Individual plasmids were re-transformed into the EGY48 strain containing the bait and reporter genes. Colonies growing on Gal(UTHL)$^-$were streaked onto selection medium (From left to right, top to bottom Gal(UTHL)$^-$, Glc(UTHL)$^-$, Gal-Xgal-(UTH)$^-$and Glc-Xgal-(UTH)$^-$).

(C) Testing for self-activation of putative interactors

Confirmed putative interactor plasmids were transformed into EGY48 containing reporter genes. Colonies growing on (UT)$^-$ medium were streaked onto Gal (UTL)$^-$ medium. The images were acquired one week after plating.