

Supplemental Data

AtLURE1/PRK6-Mediated Signaling Promotes Conspecific Micropylar Pollen Tube Guidance

Meiling Liu^{1,†}, Zhijuan Wang^{1,†}, Saiying Hou¹, Lele Wang², Qingpei Huang¹, Hongya Gu^{1,3}, Thomas Dresselhaus², Sheng Zhong^{1,*}, and Li-Jia Qu^{1,3,*}

¹State Key Laboratory for Protein and Plant Gene Research, Peking-Tsinghua Center for Life Sciences at the College of Life Sciences, Peking University, Beijing 100871, People's Republic of China.

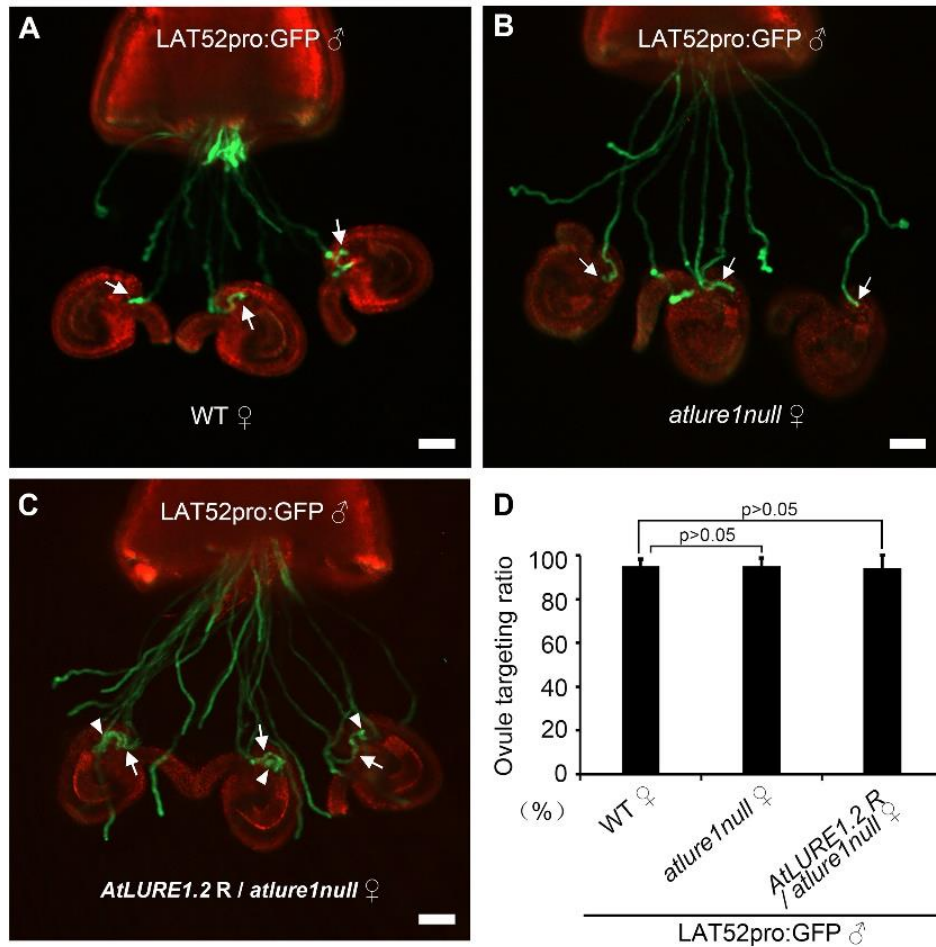
²Cell Biology and Plant Biochemistry, University of Regensburg, 93053 Regensburg, Germany.

³The National Plant Gene Research Center (Beijing), Beijing 100101, People's Republic of China.

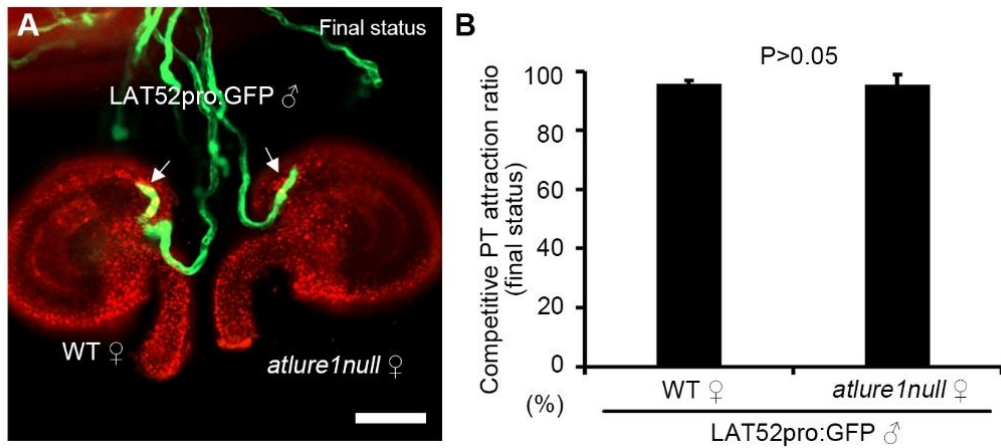
[†]The authors equally contribute to the work.

*Senior authors. Emails: shengshengzz@pku.edu.cn (S.Z.) and qulj@pku.edu.cn (L.-J.Q.)

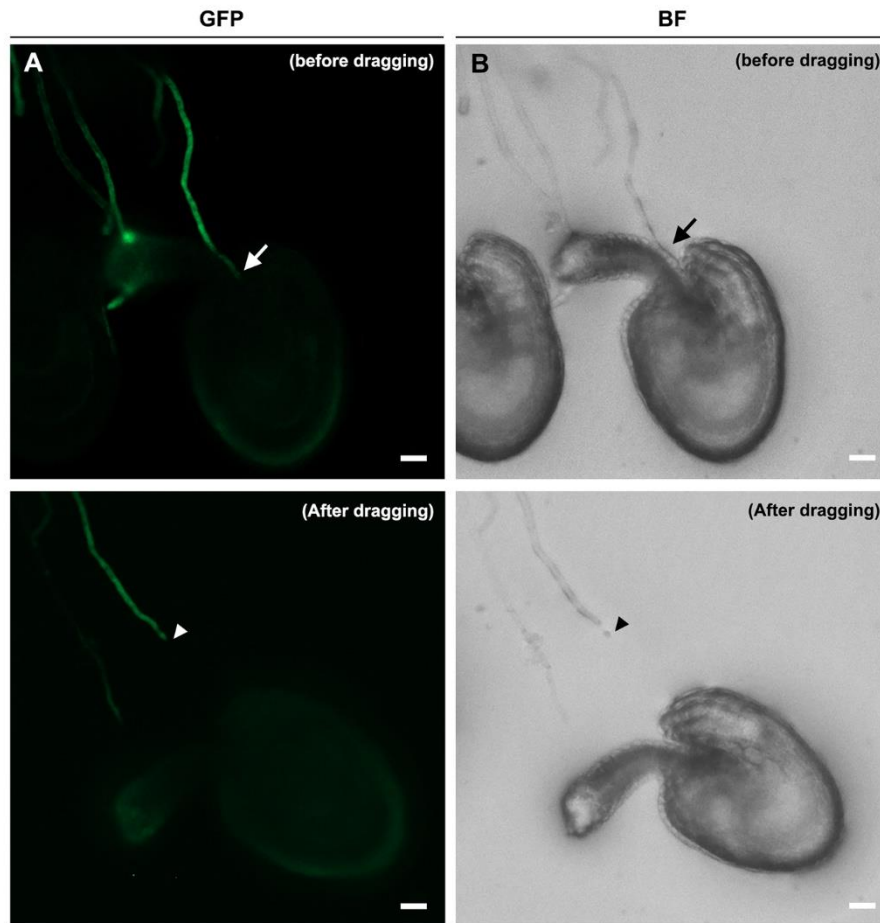
Author for correspondence: Li-Jia Qu (qulj@pku.edu.cn)



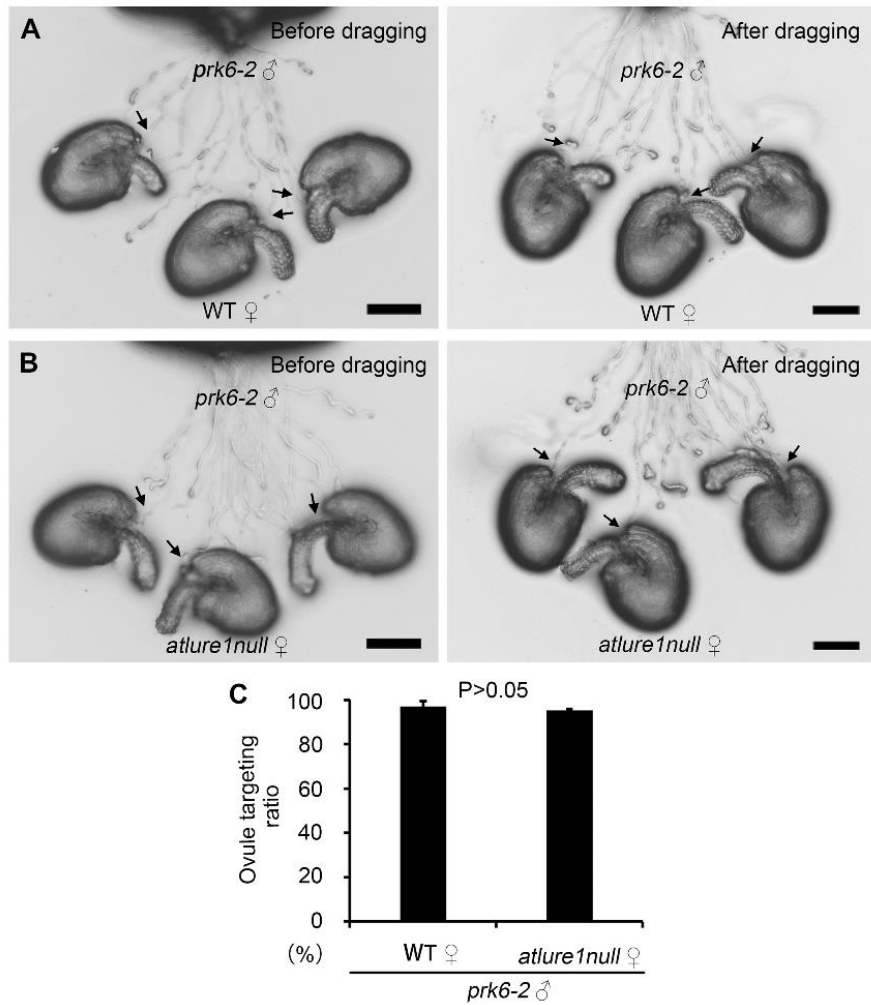
Supplemental Figure S1. LAT52pro:GFP pollen tube targeting efficiencies to WT and *atlure1null* mutant and *AtLURE1.2* rescued *atlure1null* mutant ovules, respectively. (A-C) Semi-*in vivo* pollen tube targeting assays of LAT52pro:GFP (WT) pollen to WT (A), *atlure1null* (B) and *AtLURE1.2* rescued *atlure1null* ovules (C), respectively. (D) Statistical analyses of (A-C). Four or five repeats of semi-*in vivo* pollen tube targeting assays with more than 20 ovules were each conducted. Data are mean values \pm SD. $P > 0.05$ represents no significant difference (Student's *t* test). White arrows indicate *A. thaliana* WT pollen tubes labeled with LAT52pro:GFP. White arrowheads indicate *AtLURE1.2* rescued *atlure1null* ovules labeled with *AtLURE1.2pro:AtLURE1.2-AtLURE1.2:GFP* (as *AtLURE1.2R / atlure1null*). Scale bars, 50 μ m.



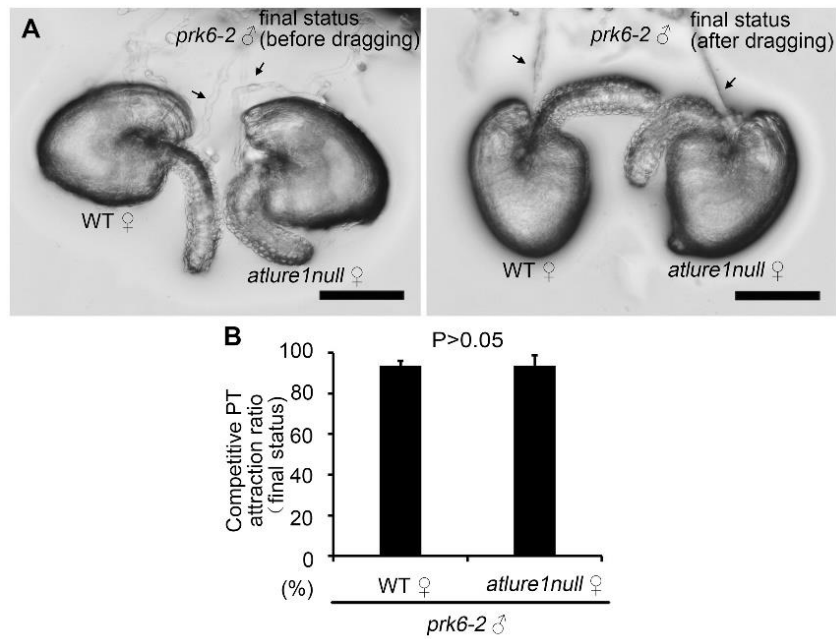
Supplemental Figure S2. Final status of WT and LAT52pro:GFP pollen tubes in the semi-*in vivo* ovule competition assay. (A) Final status showing that LAT52pro:GFP pollen tubes entered both, WT and *atlure1null* ovules. (B) Statistical analyses of (A). Four repeats of semi-*in vivo* ovule targeting assays with 20-30 ovules each were conducted. White arrows indicate *A. thaliana* WT pollen tubes labeled with LAT52pro:GFP. Data are mean values \pm SD. $P > 0.05$ means no significant difference (Student's *t* test). Scale bars, 50 μ m.



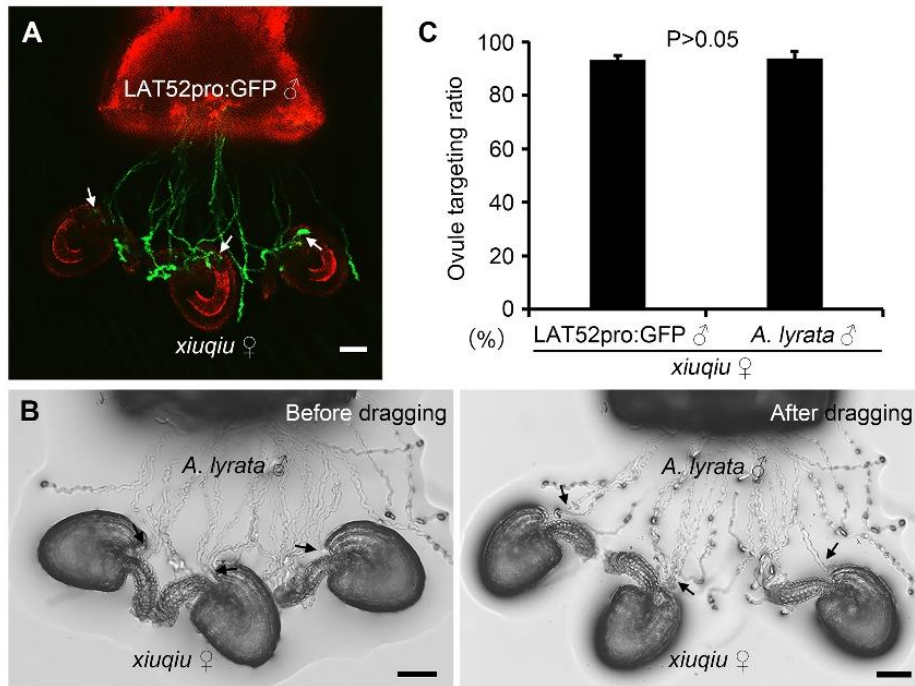
Supplemental Figure S3. Estimation of the targeting status in GFP channel and bright field. (A and B) Semi-*in vivo* pollen tube targeting assay with WT ovules targeted by LAT52pro:GFP labeled pollen tubes in the GFP channel (A) and by using bright field microscopy (B). GFP signals after dragging show that LAT52pro:GFP labeled pollen tube did not target the ovule. Arrows indicate LAT52pro:GFP labeled pollen tubes and arrowheads point towards the intact pollen tube tip. Note that in the bright field image before dragging it seems that the pollen tube targets the ovule, but dragging demonstrates that the pollen tube did not enter the micropyle. Scale bars, 50 μm .



Supplemental Figure S4. *prk6-2* mutant pollen tube targeting efficiencies to WT and *atlure1null* mutant ovules, respectively. (A and B) Bright field images of semi-*in vivo* pollen tube targeting assay with WT and *atlure1null* mutant ovules targeted by *prk6-2* pollen tubes before and after dragging as indicated. (C) Statistical analyses of (A and B). Four repeats of semi-*in vivo* ovule targeting assays with more than 20-30 ovules were each conducted. Data are mean values \pm SD. $P > 0.05$ represents no significant difference (Student's *t* test). Black arrows indicate *prk6-2* mutant pollen tubes. Scale bars, 50 μ m.



Supplemental Figure S5. Confirmation of the final status of *prk6-2* pollen tubes in the semi-*in vivo* ovule competition assay by “dragging” method. (A) Final status showing that *prk6-2* pollen tubes entered both, WT and *atlure1null* ovules. Left image before dragging and right image after dragging. (B) Statistical analyses of (A). Four repeats of semi-*in vivo* ovule targeting assays with more than 20-35 ovules each were conducted. Black arrows indicate *prk6-2* mutant pollen tubes. Data are mean values \pm SD. $P > 0.05$ means no significant difference (Student’s *t* test). Scale bars, 50 μm .



Supplemental Figure S6. Pollen tube targeting efficiencies of LAT52pro:GFP pollen tube and *A. lyrata* pollen tube to *xiuqiu* mutant ovules, respectively. (A) Semi-*in vivo* pollen tube targeting assays of LAT52pro:GFP pollen tube towards *xiuqiu* mutant ovules. (B) Semi-*in vivo* pollen tube targeting assays of *A. lyrata* pollen tube to *xiuqiu* ovules before and after dragging as indicated. (C) Statistical analyses of (A) and (B). Three repeats of semi-*in vivo* pollen tube targeting assays with 30-60 ovules were conducted each. Data are mean values \pm SD. $P > 0.05$ represents no significant difference (Student's *t* test). White arrows indicate *A. thaliana* WT pollen tubes labeled with LAT52pro:GFP. Black arrows indicate *A. lyrata* pollen tubes. Scale bars, 50 μ m.