FAFU-UCR HBMC

Self-promoted and self-terminated PIN2-mediated auxin transport is crucial during coordination of pavement cell (PC) morphogenesis

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INTRODUCTION

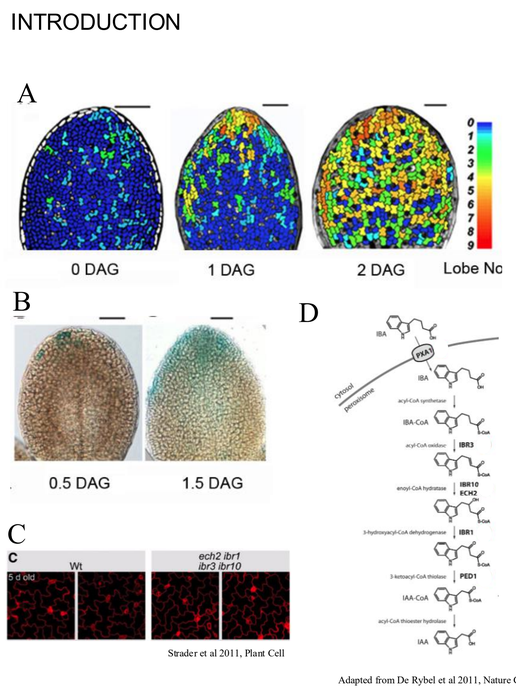


Figure 1. Progressive tip-to-base activation of pavement cell (PC) interdigitation is preceded by tip-high auxin transcriptional response.
(A) Progression of Pavement cell morphogenesis (interdigitation) occurs gradually over time within 48 h
(B) Auxin transcriptional response showed highest level before initiation of PC morphogenesis
(C) IBA to IAA conversion guided by enoyl-coA hydratase enzymes and paralogues is crucial for PC morphogenesis
(D) IBA to IAA conversion pathway. Notice key enzyme enoyl0coA hydratase, ECH2. The conversion occurs within peroxisomes.

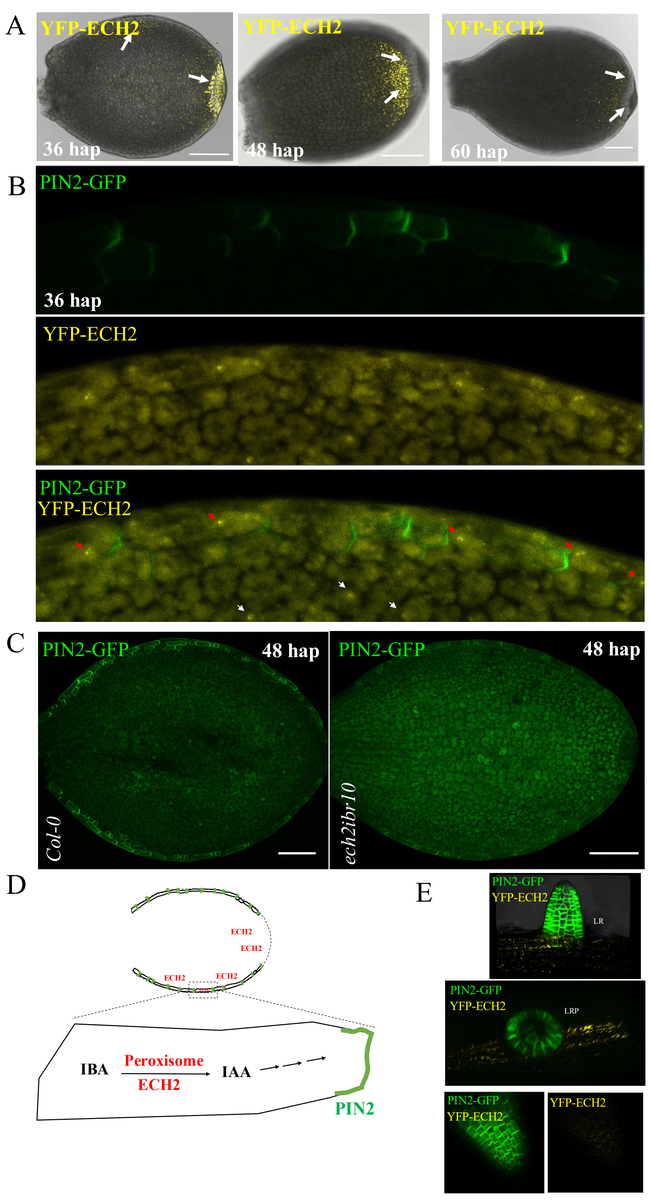


Figure 4. **IBA-derived auxin regulates PIN2 accumulation along cotyledon's margin cells.**
(**A**) YFP-ECH2 accumulation in the borders and tip of expanding cotyledon. (**B**) YFP-ECH2 accumulates at MCs suggesting IBA→IAA conversion leading to PIN2-GFP accumulation (**C**) IBA-to-IAAA conversion double mutant *ech2 ibr10* showed diminished PIN2-GFP levels at MCs (**D**) Scheme for relationship between IBA/IAA and PIN2 levels (**E**) ECH2 accumulates in other developing /expanding organs

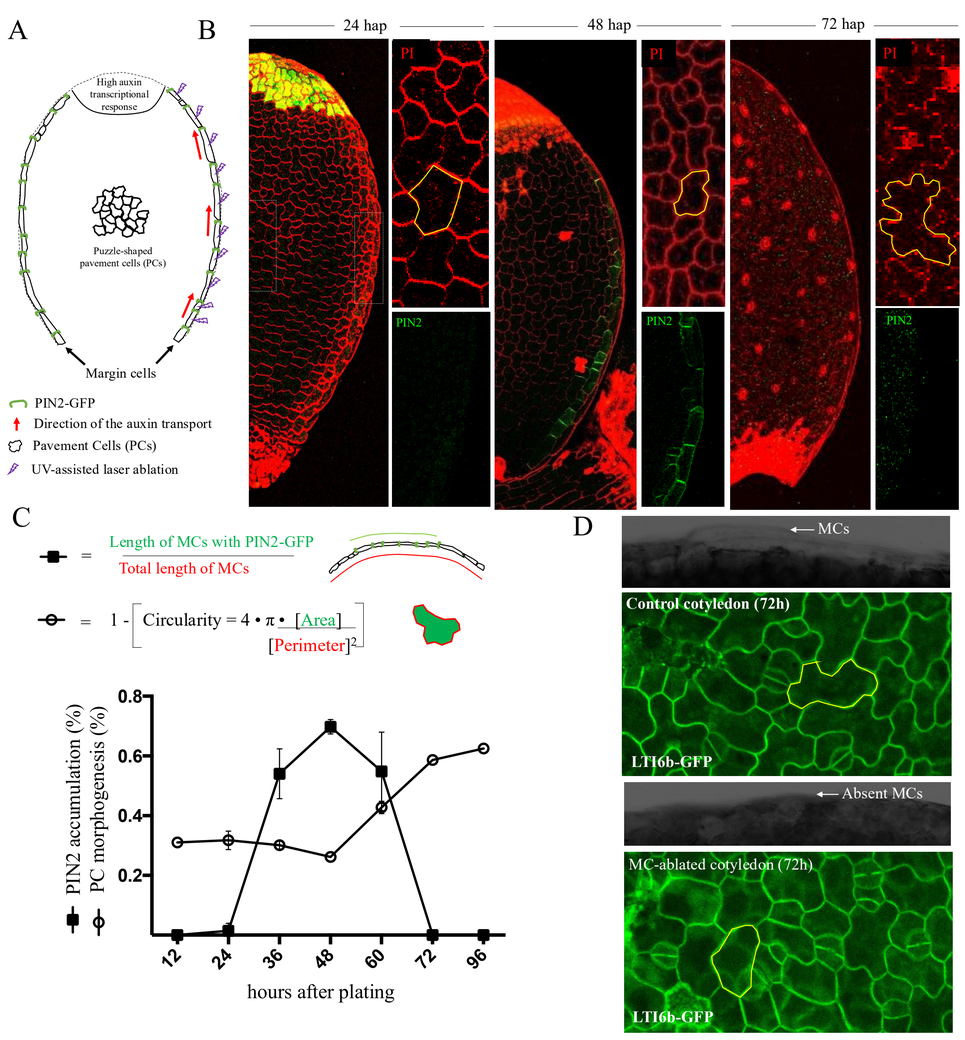


Figure 2. PIN2 accumulation in MCs precedes PC morphogenesis and its function is crucial during global coordination of pavement cell morphogenesis
(**A**) Scheme of cotyledon. (**B**) Time lapse identifying PIN2-GFP accumulation along the MCs and the progression of pavement cell morphogenesis (**C**) Quantification of PIN2 accumulation along cotyledon's margin cells (MCs) (**D**) Pavement cells when margin cell are laser-assisted ablated (**E**) Circularity of pavement cells at the tip of the cotyledon of pin2 mutants (**F**) Quantification of images in E.

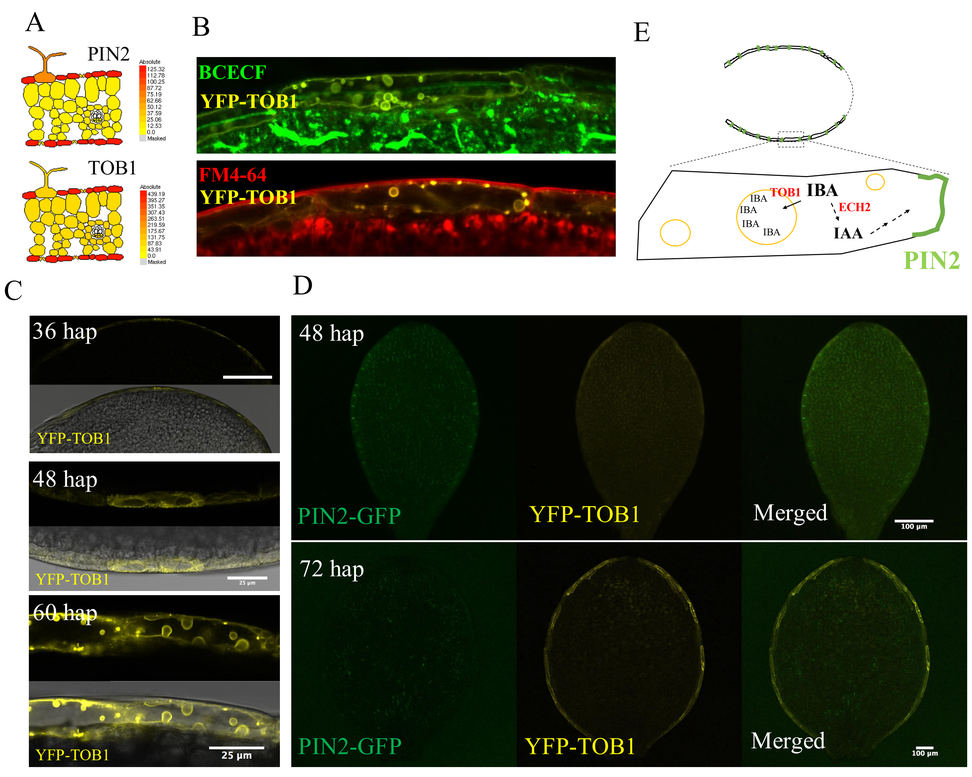
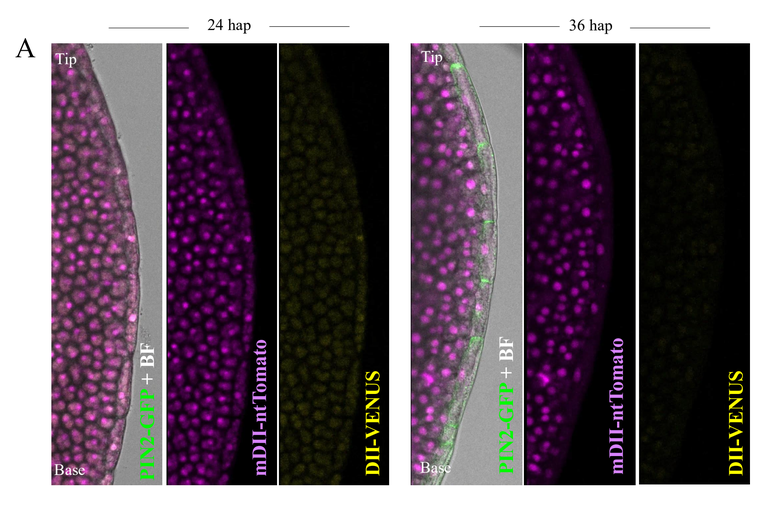


Figure 5. TOB1-mediated vacuole internalization of IBA terminates with PIN2 polar accumulation at cotyledon's margin cells.
(**A**) Expression levels for IBA transporter TOB1 is high in epidermis, similar to PIN2 (data from http://efp.ucr.edu/. (**B**) YFP-TOB1 accumulates in vacuoles within MCs according to tonoplast marker FM464 and vacuolar lumen marker BCECF (**C**) YFP-TOB1 consolidates vacuolar localization towards 60 hap (**D**) YFP-TOB1 and PIN2-GFP accumulation temporal patters correlates inversely. Together this evidence suggest that vacuolar TOB1 contributes to reduce available cytoplasmic IBA which reduces PIN2-levels



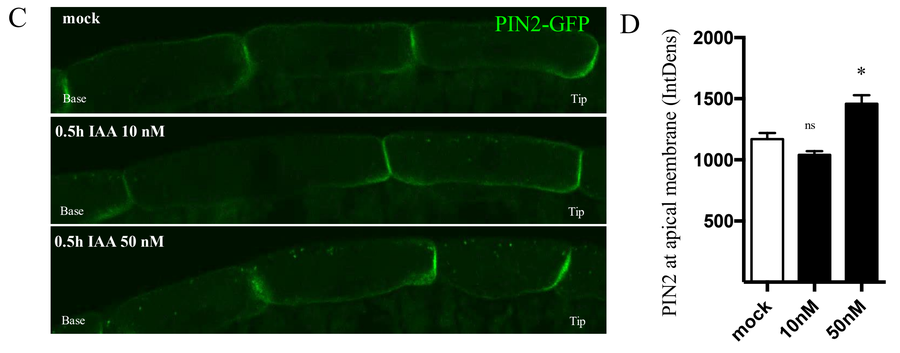


Figure 3. Auxin promotes its own PIN2-mediated efflux along cotyledon's margin cells.
(**A**) Auxin signaling reporter indicate increased auxin signaling after PIN2-GFP accumulation (**B**) Quantification of ratiometric R2D2 signal in the margin cells (**C**) PIN2-GFP levels at the apical membrane after 30 min of exogenously applied auxin IAA (**D**) Quantification of PIN2 at the apical membrane of MC cells.

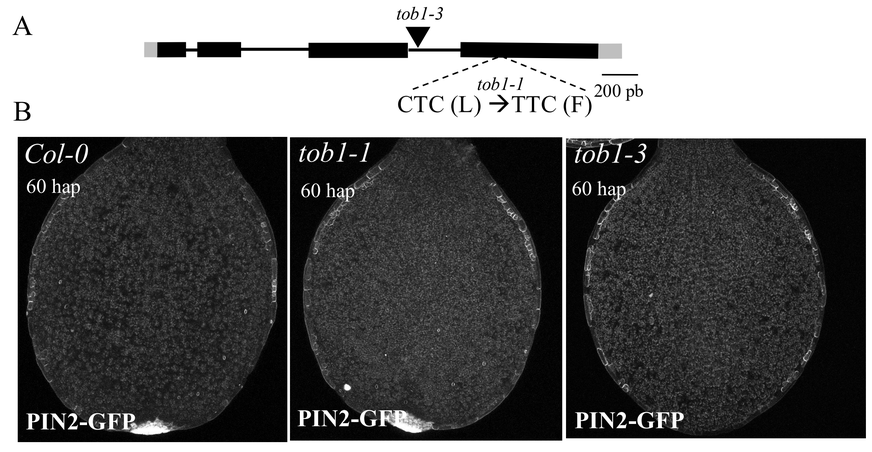
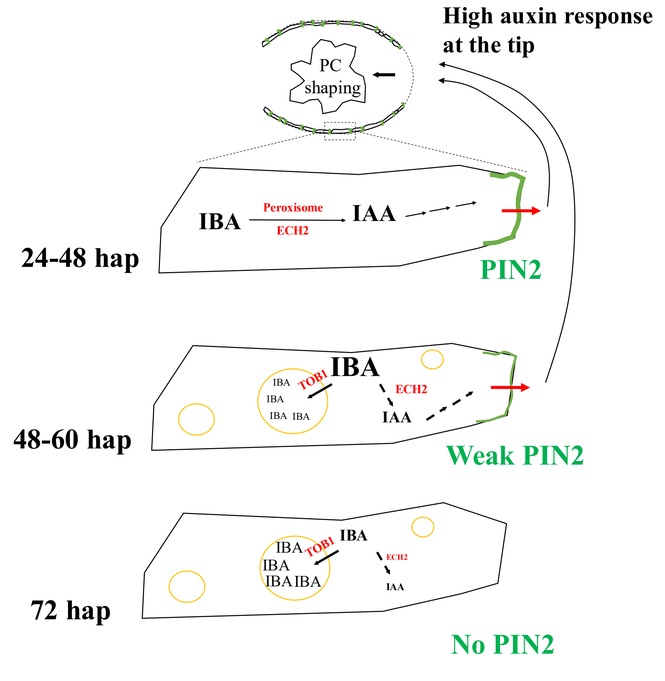


Figure 6. TOB1 function to downregulate PIN2 levels.
(**A**) Gene model for TOB1 depicting tob1-1 point mutation and tob1-3 insertional mutation(**B**) loss-of-function mutants *tob1-1* and *tob1-3* cause increased PIN2-GFP accumulation on MCs (**C**) *tob1-3* showed increased PIN2-accumulation at root tips (**D**) Quantification of PIN2 level in Col-0 and *tob1-3* background.

MODEL



CONCLUSION

1. Auxin transporter PIN2 accumulates transiently at cotyledon's margin cells preceding PC morphogenesis
2. PIN2 functions to mobilize auxin self-promoting its own transport towards the tip of the cotyledon and its function is critical for PC morphogenesis.
3. IBA-derived auxin, through ECH2 enzyme, is critical in determining PIN2 levels and PC morphogenesis
4. TOB1 is an IBA transporter that reduces cytoplasmic IBA after internalizing IBA into vacuoles.

ACKNOWLEDGMENTS

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REFERENCES

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