

# Intrinsic Control of Precise Dendritic Targeting by an Ensemble of Transcription Factors

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## Supplemental Results

### Analysis of *sqz* in PN Dendritic Targeting

*squeeze* encodes a zinc-finger TF and has been shown to regulate neuropeptide identity and axon targeting of embryonic neurons in the central nervous system [S1, S2]. Because *Sqz* antibody has not been reported, we used *sqz<sup>lacZ</sup>* [S1] to test its expression in PNs. We detected ubiquitous  $\beta$ -Gal staining in the pupal central nervous system at 18 hr APF including PNs (data not shown). It remains to be verified whether this pattern reflects the endogenous *sqz* expression.

vPN and adPN clones homozygous for the null allele (*sqz<sup>o</sup>*) exhibited normal dendritic targeting. However, *sqz<sup>-/-</sup>* IPNs showed a very specific defect. DM5 was always uninnervated, while all other glomeruli were correctly targeted (Figure S1A,  $n = 7$ ). Therefore, of the 17 classes examined, *sqz* is required for targeting of a single IPN class. This defect was rescued in at least 4 out of 9 *sqz<sup>-/-</sup>* IPN clones that expressed a UAS-*sqz* transgene (Figure S1B). Because GH146 is expressed only in postmitotic PNs (J. Liu, M. Spletter, and L.L., unpublished observation), this rescue experiment indicates that *sqz* controls targeting of DM5 IPNs postmitotically.

In contrast to *cut* and *drifter* (see Figure 4), overexpression of *sqz* in WT DL1 adPNs (Figure S1D,  $n = 10$ ; compared with Figure S1C) or *acj6<sup>-/-</sup>* DL1 adPNs (Figure S1F,  $n = 4$ ; compared with Figure S1E) did not have an effect on their dendritic targeting. The rescue experiment (Figure S1B) confirms that the transgene is

functional. Therefore, at least in DL1 single-cell clones, *sqz* does not provide instructive information on PN dendritic targeting.

### Supplemental References

- S1. Allan, D.W., St Pierre, S.E., Miguel-Aliaga, I., and Thor, S. (2003). Specification of neuropeptide cell identity by the integration of retrograde BMP signaling and a combinatorial transcription factor code. *Cell* 113, 73–86.
- S2. Allan, D.W., Park, D., St Pierre, S.E., Taghert, P.H., and Thor, S. (2005). Regulators acting in combinatorial codes also act independently in single differentiating neurons. *Neuron* 45, 689–700.

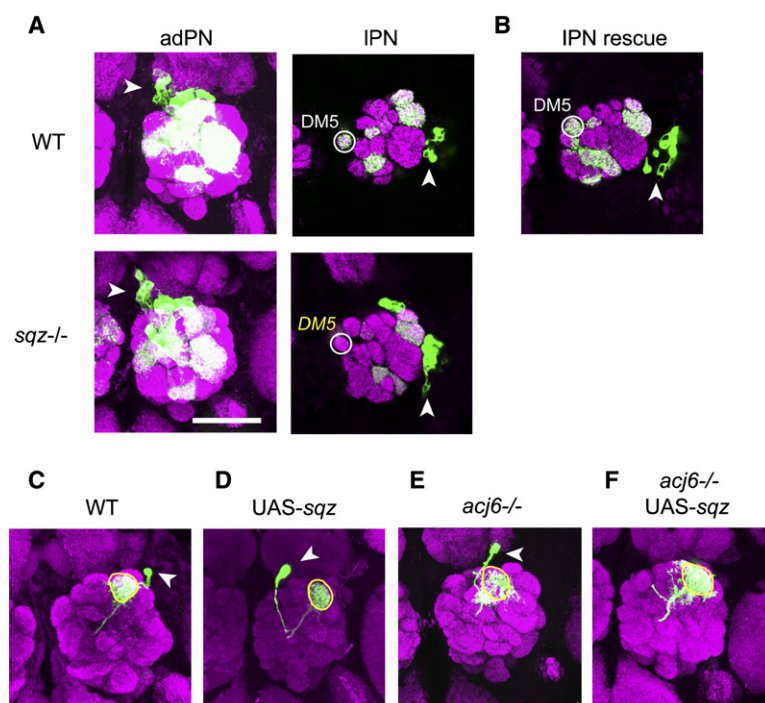


Figure S1. *sqz* Is Required for Dendritic Targeting by DM5 IPNs

(A) *sqz<sup>-/-</sup>* adPNs target all landmark glomeruli normally, but *sqz<sup>-/-</sup>* IPNs fail to target one of their target glomeruli, DM5. adPN, confocal z-projections; IPN, partial confocal z-projections.

(B) Failure of DM5 targeting by *sqz<sup>-/-</sup>* IPNs was rescued by clonal expression of a *sqz* transgene. A partial confocal z-projection is shown.

(C) WT DL1 single-cell clone.

(D) *sqz* overexpression does not alter dendritic targeting of the DL1 single-cell clone.

(E) Loss of *acj6* causes dendrites to be diffuse in a nondirectional way, but DL1 is still at least partially innervated.

(F) *sqz* overexpression in *acj6<sup>-/-</sup>* single-cell clones does not further alter their dendritic targeting.

Green, mCD8-GFP marking MARCM clones; magenta, nc82; scale bar represents 50  $\mu$ m; arrowheads, PN cell bodies.