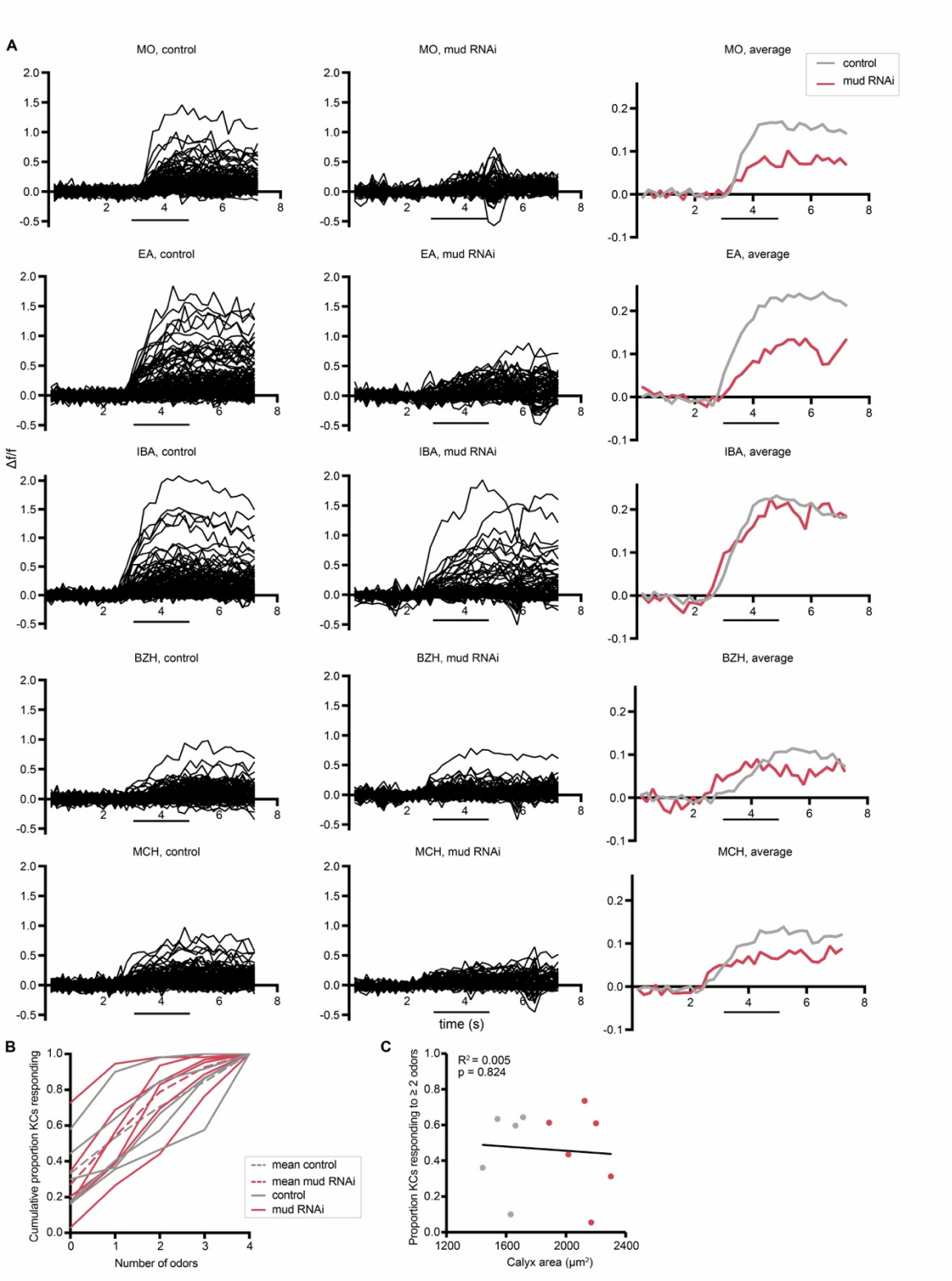

Figure S1. Supplemental data related to Figure 1.
(A) Odor responses over time for Kenyon cells shown in Figure 1G. x axes show seconds, y axes show Δf/f. Black bars indicate odor delivery. Each black line is one cell, with graphs at right showing responses averaged across all cells of the sample. Each cell was normalized to average fluorescence in the 3 s period before stimulus onset. MO: Mineral oil (mechanosensory and solvent control), EA: Ethyl Acetate, IBA: Isobutyl Acetate, BZH: Benzaldehyde, MCH: Methylcyclohexanol. An example of a motion artifact can be seen in 'MO, mud RNAi' trace around 5s. n = 124 cells (control), 62 cells (*mud* RNAi). (B) Cumulative proportion of Kenyon cells responding from 0 up to 4 odors. Each line represents an individual control hemisphere (gray) or increased-KC *mud* RNAi hemisphere (red), with the mean of all control or *mud* RNAi samples shown with a dotted line. (C) Relation of proportion of KCs responding to 2 or more odors, and maximum cross-sectional calyx area of control (gray) and KC-increased *mud* RNAi (red) hemispheres. Here and throughout, linear regressions are performed across all data points shown in the figure, i.e., for the distribution of the two sample types taken together across the variation in calyx size.

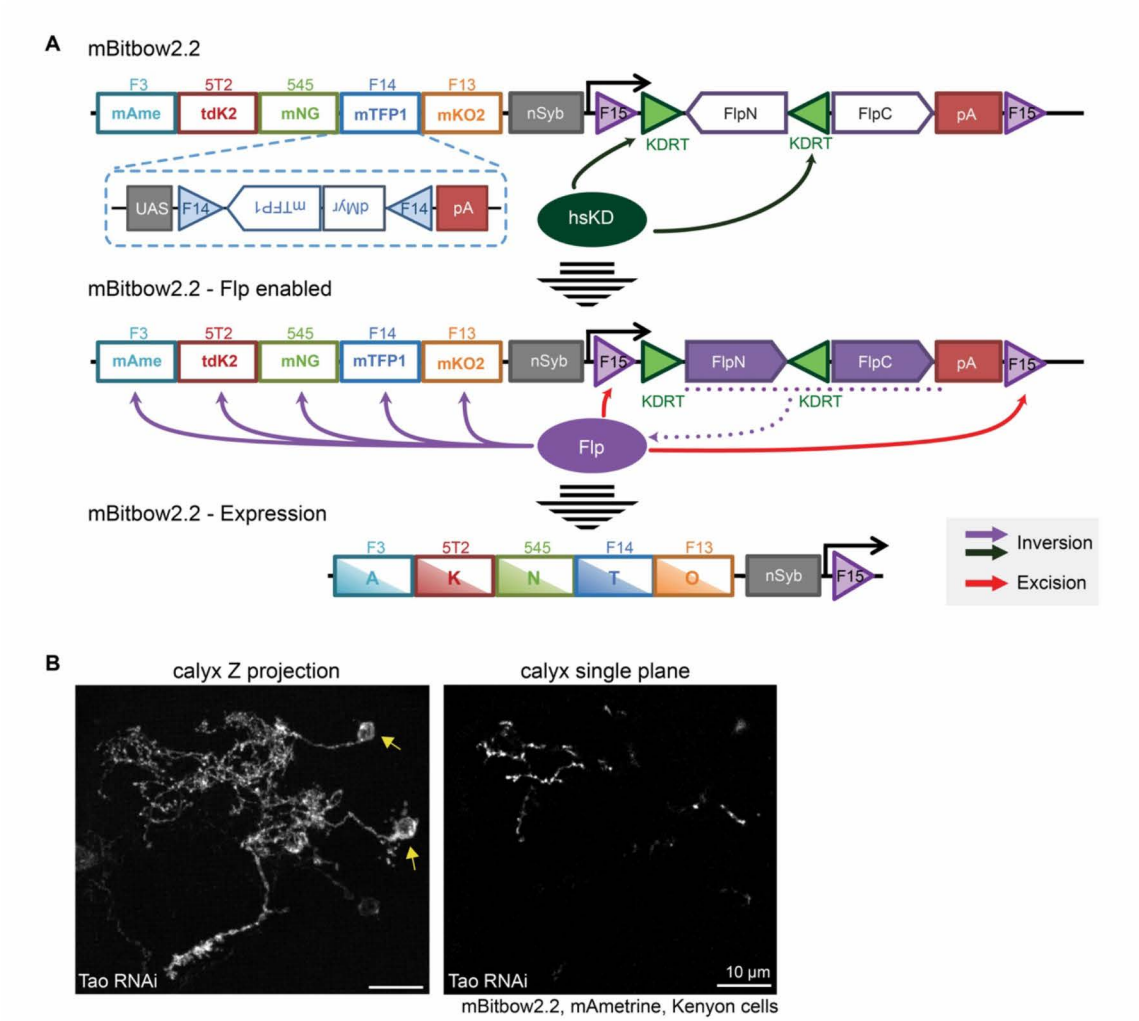

Figure S2. mBitbow2.2 schematic and dendritic morphology in *Tao*-knockdown Kenyon cells.
(A) Detailed schematic of mBitbow2.2 design; a simplified version is shown in Figure 2A. (B) Images of sparsely labeled KCs in a KC>*Tao* RNAi calyx showing highly branched but non-clawed dendritic projections that we observe in some samples: maximum z projection (left), single confocal slice (right). In the example shown, arrows mark 2 KCs labeled by mBitbow2.2 mAmetrine. Faint third soma is labeled by an additional mBitbow2.2 color.

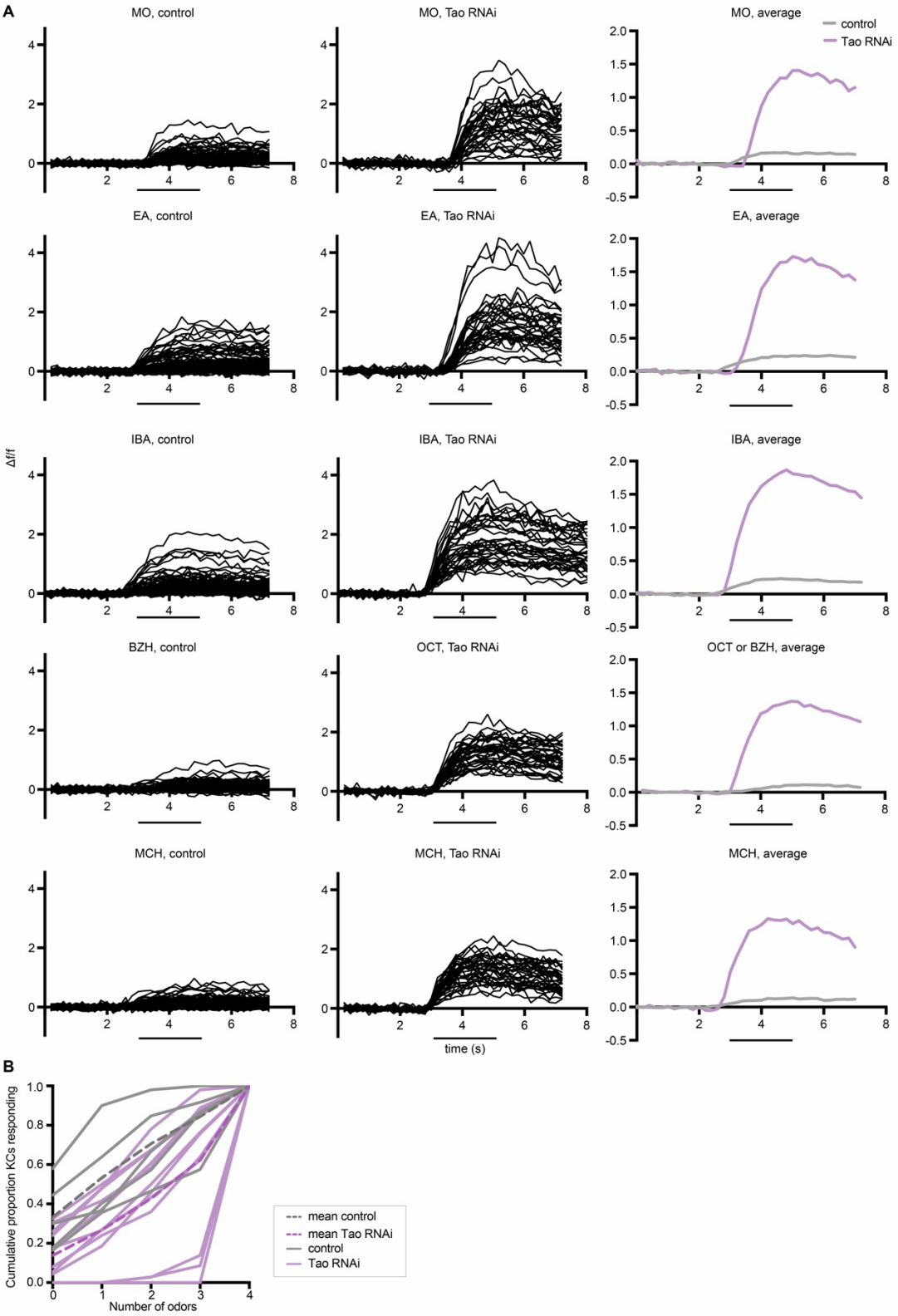

Figure S3. Odor responses over time for Kenyon cells shown in Figure 3A.

(A) x axes show seconds, y axes show Δf/f. Black bars indicate odor delivery. Each black line is one cell, with graphs at right showing responses averaged across all cells of the sample. Each cell was normalized to average fluorescence in the 3 s period before stimulus onset. MO: Mineral oil (mechanosensory control), EA: Ethyl Acetate, IBA: Isobutyl Acetate, BZH: benzaldehyde, OCT: Octanol, MCH: Methylcyclohexanol. Control sample is the same sample shown in Figure S1; we have replotted the data to allow quantitative comparison with the robust responses of excess-claw KCs. n = 124 cells (control), 36 cells (*Tao* RNAi). (B) Cumulative proportion of Kenyon cells responding from 0 up to 4 odors. Each line represents an individual control hemisphere (gray) or *Tao* RNAi hemisphere (purple), with the mean of all control or *Tao* RNAi samples shown with a dotted line.

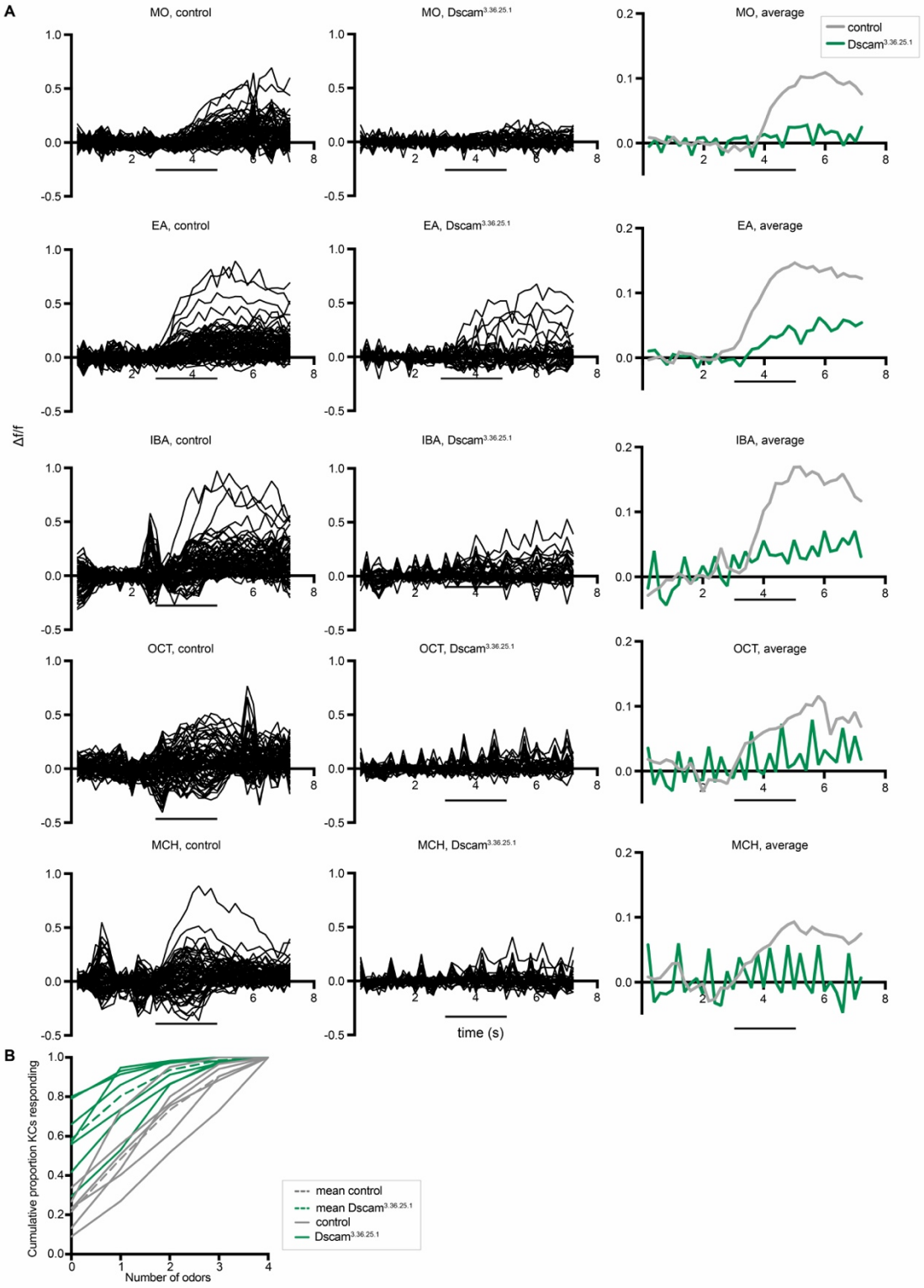

Figure S5. Odor responses over time for Kenyon cells shown in Figure 5A.

(A) x axes show seconds, y axes show Δf/f. Black bars indicate odor delivery. Each black line is one cell, with graphs at right showing responses averaged across all cells of the sample. Each cell was normalized to average fluorescence in the 3 s period before stimulus onset. MO: Mineral oil (mechanosensory control), EA: Ethyl Acetate, IBA: Isobutyl Acetate, OCT: Octanol, MCH: Methylcyclohexanol. n = 72 cells (control), 35 cells (Dscam$^{3.36.25.1}$). (B) Cumulative proportion of Kenyon cells responding from 0 up to 4 odors. Each line represents an individual control hemisphere (gray) or Dscam$^{3.36.25.1}$ hemisphere (green), with the mean of all control or Dscam$^{3.36.25.1}$ samples shown with a dotted line.

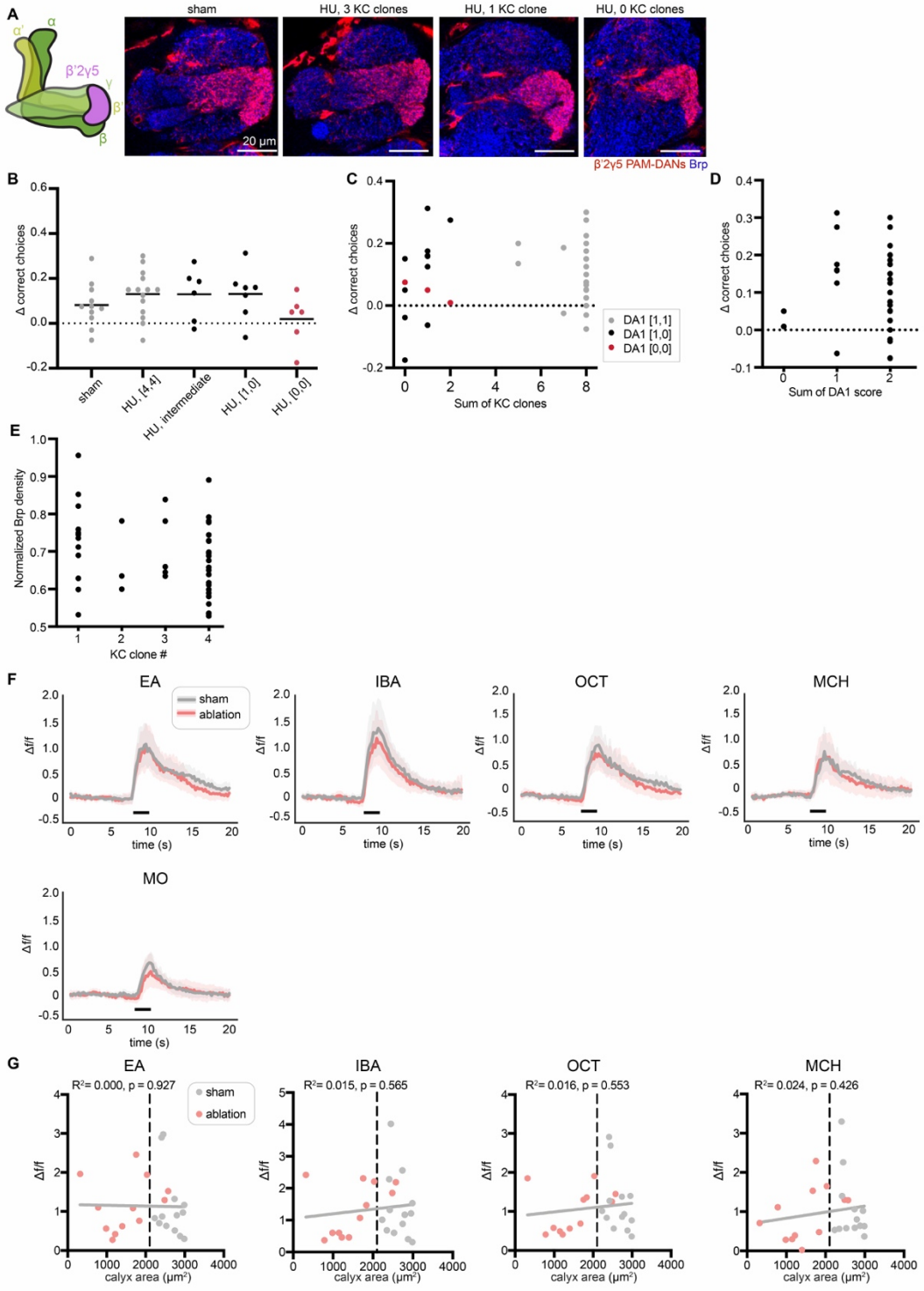

Figure S6. Supplemental data related to Figure 6

(A) Left: Schematic of the mushroom body lobe anatomy with KCs in green and β′2γ5 PAM-DANs in magenta. Axons of β′2γ5 DANs in the lobe compartment are shown. Right: Single confocal slices of the MB lobe (identified by location and Brp staining shown in blue). Mz19 driver labels β′2γ5 DANs (red). Representative images shown of sham-treated and HU-treated hemispheres with 3, 1, or 0 KC clones. (B) Δ correct choices of sham-treated and HU-treated animals shown in Figure 6D. Black bars indicate the medians. In B-D, each data point is an individual fly. (C) Relation of Δ correct choices, sum of KC clone number from both hemispheres and lNB/BAlc ablation status. DA1 present in both hemispheres is indicated as "DA1 [1,1]", presence in one hemisphere is indicated as "DA1 [1,0]", and absent in both hemispheres is indicated as "DA1 [0,0]" . (D) Relation of Δ correct choices to sum of DA1 score (presence/absence). The data shown excludes fully KC ablated animals. Jitter added in (C-E) to display all the data points. (E) Relation of normalized Brp density in the mushroom body calyx to Kenyon cell clone number in sham-treated and HU-treated animals, excluding fully ablated animals as there is no calyx present. (F) Average odor responses over time for γ2,α′1 MBONs in Figure 6I. x axes show seconds, y axes show Δf/f. Black bars indicate odor delivery. Shadows are 95% confidence intervals for corresponding averaged traces. Each cell was normalized to average fluorescence in the 5 s to 2 s period before stimulus onset. MO: Mineral oil (mechanosensory control), EA: Ethyl Acetate, IBA: Isobutyl Acetate, OCT: Octanol, MCH: Methylcyclohexanol. n= 12 hemispheres (sham), 10 hemispheres (ablation). Only HU-partially ablated hemispheres smaller than every control (maximum cross-sectional calyx area < 2100 μm^2) are included. This cutoff is labeled as black vertical dashed line in (G). (G) Relationship between γ2,α′1 MBON peak odor responses and maximum cross-sectional calyx area. Gray line is linear regression for all samples. n= 12 hemispheres (sham), 12 hemispheres (ablation). 2 HU-treated animals with maximum cross-sectional calyx area > 2100 μm^2 are also shown.

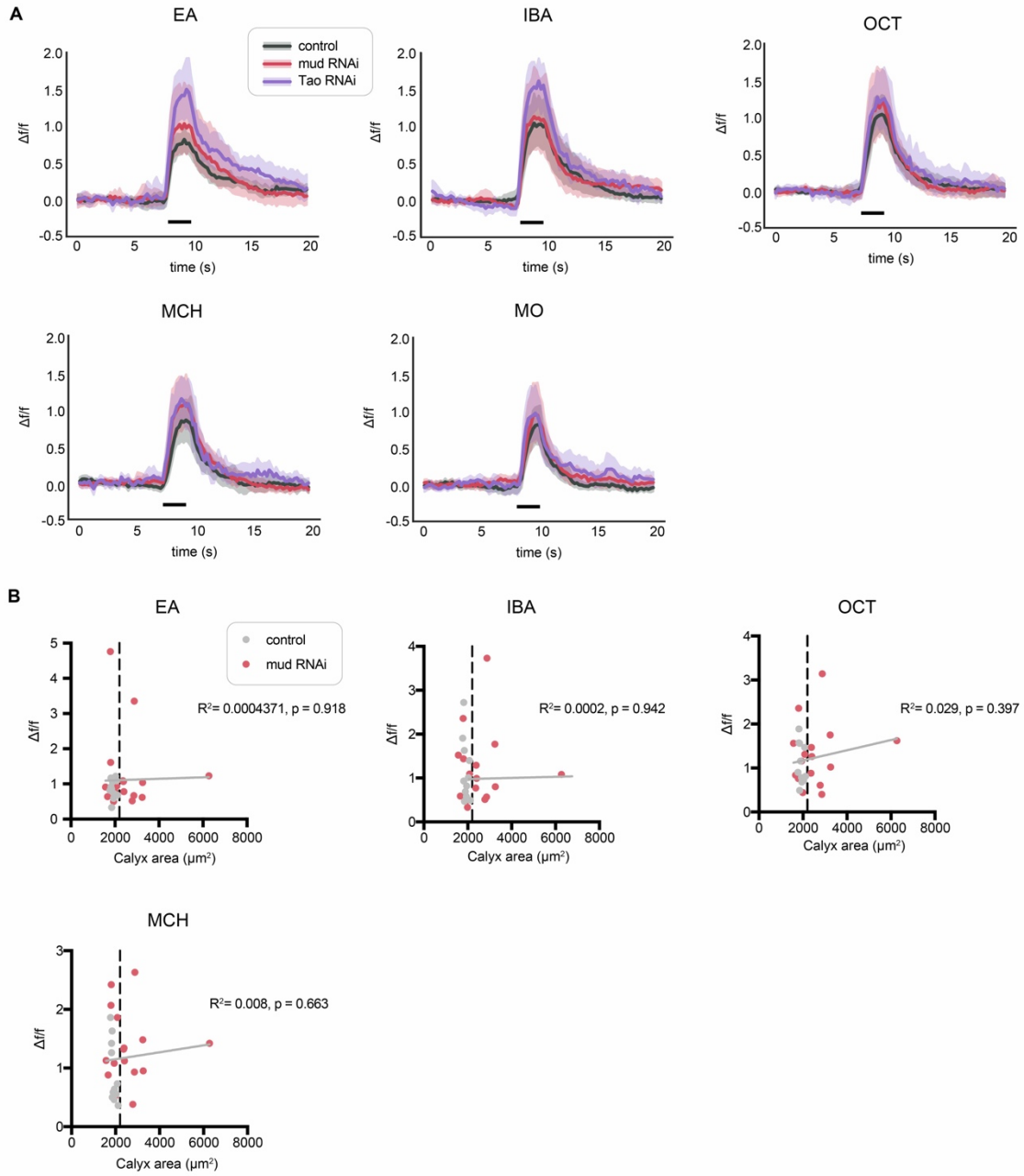

Figure S7. Odor responses for γ2,α′1 MBON in *mud* RNAi and *Tao* RNAi animals.
(A) Average odor responses over time for γ2,α′1 MBONs shown in Figure 7G. x axes show seconds, y axes show Δf/f. Black bars indicate odor delivery. Shadows are 95% confidence intervals for corresponding average trace. Each cell was normalized to average fluorescence in the 5 s to 2 s period before stimulus onset. MO: Mineral oil (mechanosensory control), EA: Ethyl Acetate, IBA: Isobutyl Acetate, OCT: Octanol, MCH: Methylcyclohexanol. n= 10 hemispheres (control), 9 hemispheres (*mud* RNAi), 12 (*Tao* RNAi). For *mud* RNAi, only Kenyon cell-increased hemispheres (maximum cross-sectional calyx area > 2200 μm^2) are included. This threshold is labeled as black vertical dashed line in (B). (B) Relationship between γ2,α′1 MBON peak odor responses and maximum cross-sectional calyx area. Gray line is linear regression for all samples. n= 10 hemispheres (control), 15 hemispheres (*mud* RNAi); six hemispheres from *mud*

RNAi calyces with calyx cross-sectional area overlapping controls ($< 2200\ \mu m^2$) are included among these 15.

Parameter	MB Name	Variable Name	Number in natural MB	Number engineered here
Number of inputs	Olfactory projection neuron types	N	52	40-52
Number of expansion layer neurons	Kenyon cells per hemisphere	M	2000	500-4000
Expansion ratio (M/N)	Kenyon cell number/odor channels	E	38	10-77
Number of inputs to each expansion layer neuron	Claw number	K	5	1-12
Spiking threshold	Number of input channels active for KC to spike	?	Usually 2 or more, occasionally 1	*We suspect it has not changed
Strength of feedback inhibition	APL activity	Sigma	Unknown	Unknown
Total connections between sensory and expansion layer	Total number of Kenyon cell claws	S	10,000	2,000-24,000

Supplemental Table 1: Quantitative variables of mushroom body calyx wiring and function

Condition	Result	N	M	E	K	KC spike threshold	Sigma	S
Wild type	KCs respond to 0-1 odors	52	2000	38	5-6	~2 claws	unknown	10,000
5HU ablation	Fewer KCs, but each responds to the same number of odors	40-52	500-2000	10-50	5-6	~2 claws (inferred)	Proportional to KCs active (inferred)	2,500-10,000
Mud knockdown	More KCs, but each responds to the same number of odors	52	2000-4000	38-77	6	~2 claws (inferred)	Proportional to KCs active (inferred)	10,000-24,000

Tao knockdown	Each KC responds to more odors	52	1700	32	12	~2 claws (inferred)	Proportional to KCs active (inferred)	~20,400
Dscam[TM1] overexpression	Each KC responds to fewer odors	52	1800	35	1	1-2 claws (inferred)	Proportional to KCs active (inferred)	1800

Supplemental Table 2: Summary of the effects of our developmental manipulations on mushroom body calyx connectivity variables