Neuron, Volume *76*

Supplemental Information

Drosophila CPEB Orb2A Mediates Memory Independent of its RNA-Binding Domain

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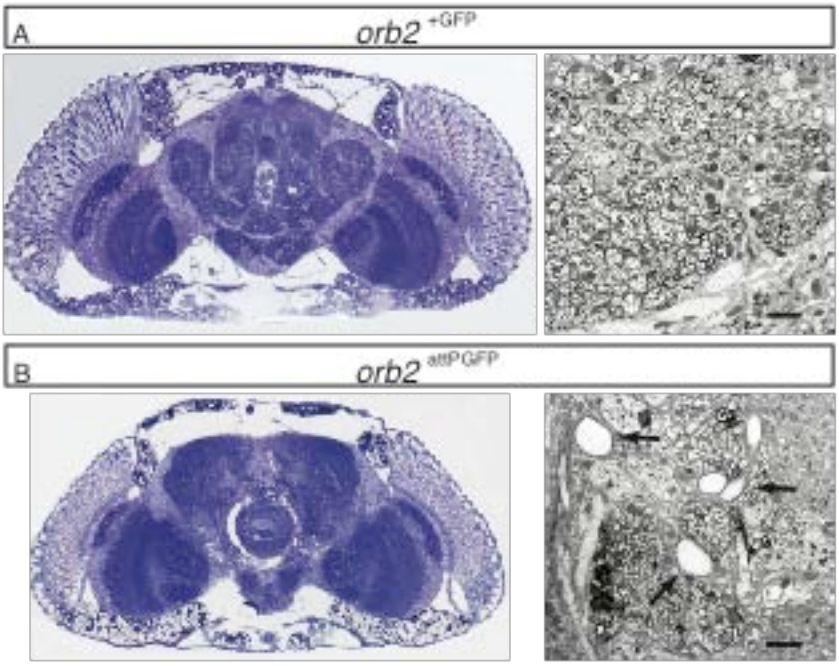
Inventory of the Supplemental Information

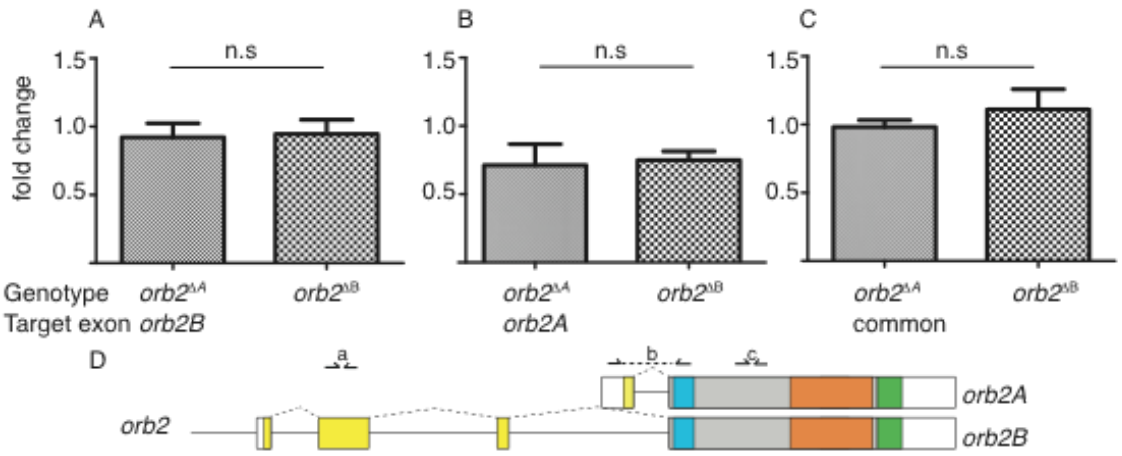
Supplemental Figures

Fig. S1 Morphological defects in the brain of *orb2*^{attPGFP} null mutants



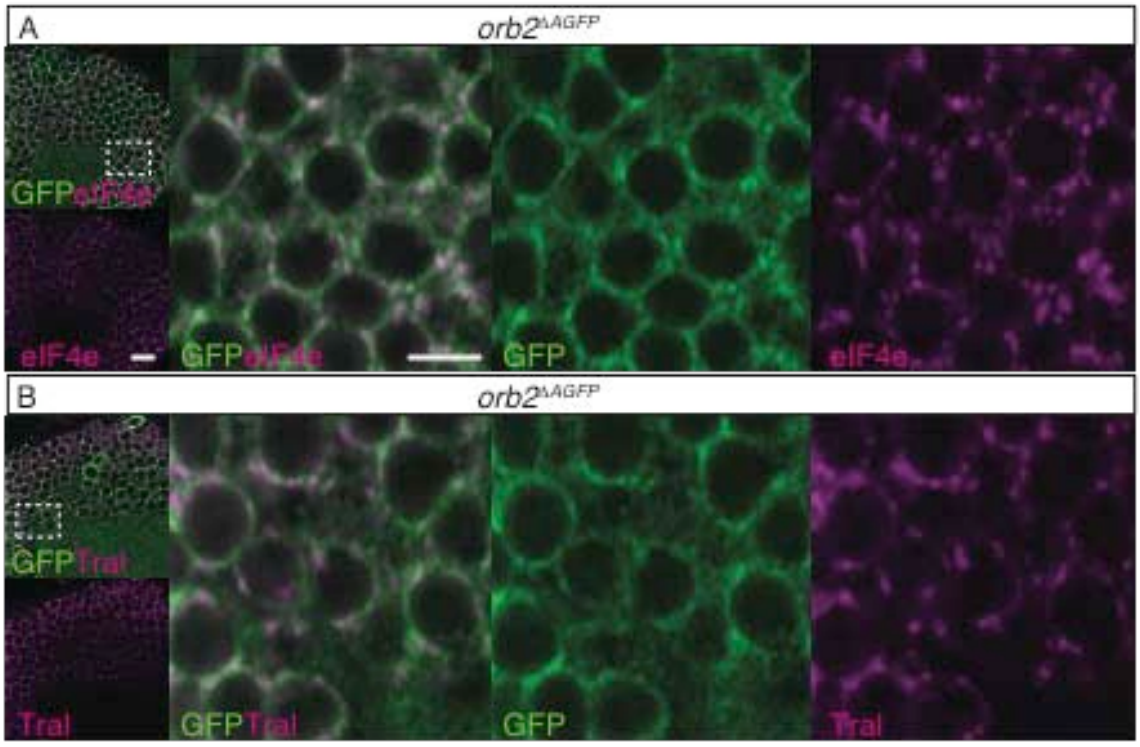
A, a toluidine blue-stained semi-thin frontal brain section showing the general organization of the brain of *orb2*$^{+GFP}$ allele (upper left panel). The general morphology resembles that of the Canton-S brains (not shown).
A micrograph of a section from the sample on the left, that shows high GFP expression in many axons in a bundle located in an area, likely the antennocerebral tract, next to the ellipsoid body (upper right panel). Scale bar is 2 um.
B, a toluidine blue-stained semi-thin frontal brain section showing the brain of *orb2*^{attPGFP} null mutant (lower left panel). The overall size of the brain is significantly reduced compared to the *orb2*$^{+GFP}$ brain.
A micrograph taken from the brain sample on the left shows a large number of vacuole-like structures (arrows) (lower right panel). These structures are rarely seen in the *orb2*$^{+GFP}$ wild type brains. Scale bar is 2 um.

Fig. S2 Deletion of either Orb2 isoform does not affect transcript levels from the *orb2* locus



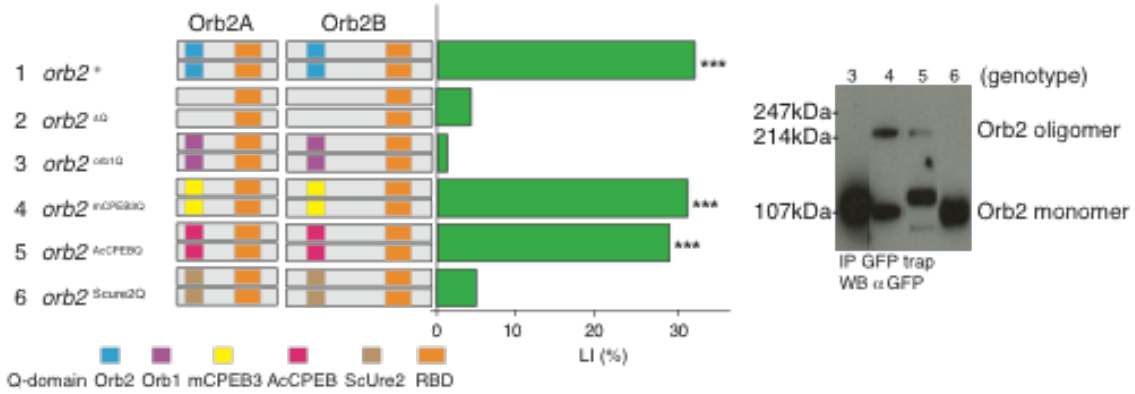
Q-PCR targeting *orb2B* specific exon **A**) *orb2A* specific exon **B**) *orb2* common exon **C**). Relative isoform specific transcript levels for indicated genotypes ± s.e.m. in comparison to *orb2*$^{+}$ (Supplementary Table S3). **D**) genomic organization of the *orb2* locus illustrating relevant transcripts as well as primer location.

Fig. S3 Orb2B is present in the RNPs



Confocal projections of the *Drosophila* Mushroom Body (cell bodies of the Kenyon Cells) stained with the antibody to GFP to visualize Orb2B^{+GFP} (green) and to either elf 4e (red) (**A**) or Tral (red) (**B**), the RNP markers. Scale bar is 10 um (small panels) and 50 um (large panels).

Fig. S4 The Q-domain of Orb2 can be substituted by the homologous domain from other species



LIs (green bars) of males carrying the indicated *orb2* alleles (left) with the corresponding Orb2A and Orb2B protein organization (middle) tested in single-pair assays with mated females as trainers and testers for long-term memory (left panel). *P* values determined by permutation test for null hypothesis H_0: LI=0 and H_0: LI=LI_1. Supplementary Table S6. Adult flies of indicated genotypes, after being starved for 16 hrs, were fed with tyramine and sucrose for 6 hrs. Head extracts were analyzed by IP and WB for Orb2 oligomers at 24 hrs (right panel).

Supplemental Tables

Table S1 Verification of the *orb2* alleles in long-term courtship learning paradigm (LTM)

	Genotype	CI$^-$ (%)	n	CI$^+$ (%)	n	LI(%)	*P LI=0*	*P LI=LI$_1$*
1	Canton-S	80.25±0.02	49	58.41±0.02	51	32.56	<0.0001	
2	*orb2*$^{+GFP}$	83.37±0.02	52	56.94±0.03	54	31.69	<0.0001	0.883
3	*orb2*$^{\Delta QGFP}$	69.04±0.03	52	68.60±0.03	50	0.63	0.476	0.001
4	*orb2*$^{\Delta QGFP}$/*orb2*$^{att P}$	68.08±0.03	60	66.02±0.03	59	3.03	0.331	0.004

Courtship indices of naive (CI$^-$) and experienced (CI$^+$) males of the indicated genotypes according to Fig. 1C, tested in single-pair assays with mated females as trainers and testers for long-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Table S2 Verification of the *orb2* alleles in short-term courtship learning paradigm (STM)

	Genotype	CI$^-$ (%)	n	CI$^+$ (%)	n	LI(%)	*P LI=0*	*P LI=LI$_1$*
1	Canton-S	78.72±0.03	43	48.22±0.04	45	38.74	<0.0001	
2	*orb2*$^{+GFP}$	84.30±0.02	50	48.60±0.02	50	42.34	<0.0001	0.728

3	$orb2^{\Delta QGFP}$	71.02±0.03	54	37.21±0.03	52	47.60	<0.0001	0.636
4	$orb2^{\Delta QGFP}/orb2^{attP}$	73.40±0.03	53	47.24±0.04	49	35.63	<0.0001	0.483

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 1C, tested in single-pair assays with mated females as trainers and testers for short-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Table S3 Deletion of either Orb2 isoform does not affect the transcript levels form the o*rb2* locus

Genotype	**Target**	**Mean ΔCt ±s.e.m**	**n**	**Fold change to *Orb2*$^+$($2^{-\Delta\Delta Ct}$)±s.e.m**	**n**	**$2^{-\Delta\Delta Ct} = 1$**	**$2^{-\Delta\Delta Ct}_{orb2\Delta A} = 2^{-\Delta\Delta Ct}_{orb2\Delta B}$**
$orb2^+$	*orb2B*	-0.289±0.01	9				
$orb2^{\Delta A}$	*orb2B*	-0.327±0.15	9	0.92±0.10	3	0.654	
$orb2^{\Delta B}$	*orb2B*	-0.394±0.12	9	0.94±0.10	3	0.525	0.879
$orb2^+$	*orb2A*	-1.542±2.01	9				
$orb2^{\Delta A}$	*orb2A*	-2.042±1.87	9	0.72±0.15	3	0.203	
$orb2^{\Delta B}$	*orb2A*	-2.278±1.67	9	0.74±0.06	3	0.061	0.303
$0rb2^+$	*orb2* com exon	-0.394±0.12	9				
$orb2^{\Delta A}$	*orb2* com exon	-0.267±0.11	9	0.98±0.04	3	0.528	
$orb2^{\Delta B}$	*orb2* com exon	-0.424±0.12	9	1.11±0.14	3	0.753	0.451

Relative gene expression analysis according to Fig. S2 for indicated genotypes ± s.e.m was performed by the comparative delta-delta CT (ΔΔCT) qPCR method. Data was normalized to the mean Ct of Actin5C, RpS8 and HSC70-4 (mean ΔCt). Mean ΔCt was used to normalize to *orb2*$^+$ ($2^{-\Delta\Delta Ct}$). *P* values determined by one-sample t test for the null hypothesis that $2^{-\Delta\Delta Ct}$ equals *orb2*$^+$ (H_0: $2^{-\Delta\Delta Ct} = 1$) or for unpaired t-test (H_0: $2^{-\Delta\Delta Ct}_{orb2\Delta A} = 2^{-\Delta\Delta Ct}_{orb2\Delta B}$).

Table S4 The Q-domain in Orb2A is both required and sufficient for long-term memory

	Genotype	**CI^- (%)**	**n**	**CI^+ (%)**	**n**	**LI(%)**	***P LI=0***	***P LI=LI_1***
1	$orb2^+$	84.53±0.02	43	58.91±0.03	46	30.31	<0.0001	
2	$orb2^{\Delta Q}$	69.59±0.03	49	68.10±0.04	42	2.15	0.381	0.007
3	$orb2^{\Delta A}$	90.37±0.01	54	78.90±0.03	50	12.69	0.0007	0.037 **0.178 (LI_3=LI_2)**
4	$orb2^{\Delta Q\Delta A}$	70.65±0.03	54	67.00±0.03	50	5.16	0.252	0.017
5	$orb2^{\Delta Q\Delta B}$/ $orb2^{\Delta A}$	79.23±0.02	52	76.96±0.02	51	2.86	0.272	0.002
6	$orb2^{\Delta B}$/ $orb2^{\Delta Q\Delta A}$	82.64±0.02	53	68.62±0.03	47	16.97	0.001	0.162 **0.57 (LI_6=LI_7)**
7	$orb2^{\Delta A}$/ $orb2^{\Delta B}$	82.22±0.02	45	65.09±0.02	54	20.83	<0.0001	0.317

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 3, tested in single-pair assays with mated females as trainers and testers

for long-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1), or as indicated.

Table S5A The RBD swap with the mCPEB2RBD rescues development but not long-term memory

	Genotype	CI^- (%)	n	CI^+ (%)	n	LI(%)	*P LI=0*	*P LI=LI₁*
1	$orb2^{+}$	79.79±0.03	47	53.94±0.04	52	32.39	0.0003	
2	$orb2^{\Delta Q}$	69.59±0.03	49	68.10±0.04	42	2.15	0.384	0.015
3	$orb2^{Orb1RBD}$	lethal						
4	$orb2^{mCPEB2RBD}$	60.10±0.02	52	56.39±0.03	54	6.16	0.203	0.035

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 4A, tested in single-pair assays with mated females as trainers and testers for long-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Table S5A' The RBD swap with the mCPEB2RBD allele has a normal short-term memory

	Genotype	CI^- (%)	n	CI^+ (%)	n	LI(%)	*P LI=0*	*P LI=LI₁*
1	$orb2^{+}$	84.30±0.02	50	48.60±0.02	50	42.34	<0.0001	
2	$orb2^{\Delta Q}$	71.02±0.03	54	37.21±0.03	52	47.60	<0.0001	0.511
3	$orb2^{mCPEB2RBD}$	78.85±0.02	52	47.31±0.02	52	40.00	<0.0001	0.736

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 4A, tested in single-pair assays with mated females as trainers and testers for short-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Table S5B RBD is essential for function of Orb2B but not of Orb2A in long-term memory

	Genotype	CI^- (%)	n	CI^+ (%)	n	LI(%)	*P LI=0*	*P LI=LI₁*
1	$orb2^{+}$	79.79±0.3	47	53.94±0.04	52	32.39	<0.0001	
2	$orb2^{\Delta A}/orb2^{\Delta B}$	82.22±0.2	45	65.09±0.02	54	20.83	<0.0001	0.309
3	$orb2^{\Delta A}/$ $orb2^{mCPEB2RBD}$	80.46±0.2	54	57.50±0.03	54	28.53	<0.0001	0.747
4	$orb2^{\Delta B}/$ $orb2^{mCPEB2RBD}$	37.77±0.3	47	37.13±0.02	54	1.68	0.430	0.039
5	$orb2^{RBD*\Delta A}/$ $orb2^{mCPEB2RBD}$	62.44±0.4	45	61.79±0.3	53	1.04	0.446	0.014
6	$orb2^{RBD*\Delta B}/$ $orb2^{\Delta A}$	84.89±0.2	47	67.41±0.03	54	20.59	<0.0001	0.287
7	$orb2^{RBD*\Delta A}/$ $orb2^{\Delta B}$	lethal						
8	$orb2^{\Delta Q\Delta A}/$ $orb2^{RBD*\Delta B}$	78.10±0.3	50	61.94±0.03	54	20.68	0.0002	0.13 **0.4** **LI_8=LI_2**

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 4B, tested in single-pair assays with mated females as trainers and testers for long-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1), or as indicated.

Table S5C RBD in Orb2B has a specific role in long-term memory in adults

	Genotype	**CI^- (%)**	**n**	**CI^+ (%)**	**n**	**LI(%)**	***P LI=0***
1	*TubG80ts, orb2*^{mCPEB2RBD}*,UAS-orb2B, MB247G4* (30°C)	79.86±0.03	35	57.21±0.04	34	28.36	0.0004
2	*TubG80ts, orb2*^{mCPEB2RBD}*,UAS-orb2B, MB247G4* (19°C)	57.03±0.05	32	52.92±0.04	36	7.21	0.282
3	*TubG80ts, orb2*^{mCPEB2RD*}*,UAS-orb2B, MB247G4* (30°C)	66.57±0.03	35	62.65±0.04	34	5.89	0.262

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 4C, tested in single-pair assays with mated females as trainers and testers for long-term memory at indicated temperatures, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0).

Table S5D orb2 alleles with the LTM defect have normal short-term memory

	Genotype	**CI^- (%)**	**n**	**CI^+ (%)**	**n**	**LI(%)**	***P LI=0***	***P LI=LI$_1$***
1	*orb2*$^+$							
2	*orb2*$^{\Delta B}$/ *orb2*^{mCPEB2RBD}	51.42±0.02	53	24.07±0.02	54	53.17	<0.0001	0.217
3	*orb2*$^{RBD*\Delta A}$/ *orb2*^{mCPEB2RBD}	57.41±0.02	54	28.70±0.02	54	50.00	<0.0001	0.370
4	*orb2*$^{\Delta A}$	88.33±0.01	54	55.46±0.03	54	37.21	<0.0001	0.442
5	*orb2*$^{\Delta Q\Delta A}$	72.71±0.02	48	36.85±0.03	54	49.31	<0.0001	0.348
6	*orb2*$^{\Delta Q\Delta B}$/ *orb2*$^{\Delta A}$	71.91±0.02	47	40.77±0.02	52	43.30	<0.0001	0.900

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. 3 and 4, tested in single-pair assays with mated females as trainers and testers for short-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Table S6 The Q-domain of Orb2 can be substituted by the homologous domain from other species

	Genotype	CI^- (%)	n	CI^+ (%)	n	LI(%)	*P LI=0*	*P LI=LI₁*
1	*Orb2*$^+$	77.55±0.03	52	53.15±0.03	54	31.42	<0.0001	
2	*orb2*$^{\Delta Q}$	72.50±0.03	36	69.56±0.02	34	4.05	0.304	0.007
3	*orb2*^{Orb1Q}	85.09±0.03	54	82.78±0.02	54	2.72	0.282	0.001
4	*orb2*^{mCPEB3Q}	76.81±0.04	47	52.23±0.02	47	31.99	<0.0001	0.981
5	*orb2*^{AcCPEBQ}	79.06±0.02	48	55.78±0.03	51	29.44	<0.0001	0.825
6	*orb2*^{UreQ}	84.23±0.02	52	80.71±0.02	42	4.174	0.140	0.0009

Courtship indices of naive (CI^-) and experienced (CI^+) males of the indicated genotypes according to Fig. S4, tested in single-pair assays with mated females as trainers and testers for long-term memory, shown as mean ± s.e.m. of *n* males. *P* values determined by permutation test for the null hypothesis that learning equals 0 (H_0: LI = 0) or for the null hypothesis that mutant and control males learn equally well (H_0: LI = LI_1).

Supplemental Experimental Procedures

Q-PCR

Q-PCR was performed as described by (Livak and Schmittgen, 2001). A Biorad iQ5 system was used for Q-PCR. Each sample was run in triplicates. An additional no-template control (reaction mixture without cDNA template) was included. After 94°C denaturation for 10 min, the reactions were cycled 40 times with 94°C denaturation for 15 s, and 60°C annealing for 1 min. The data was exported as .cvs and analyzed using Excel and GraphPAD Prism 5. Relative quantification was performed using the comparative threshold (CT) method ($\Delta\Delta$CT) after determining the CT values for reference (Hsp24, Act5C, HSP70-4) and target genes (*orb2B* and *orb2A* specific exons and *orb2* common exon) (Applied Biosystems; User Bulletin 2). Fold changes in target mRNA expression level were calculated after normalization to references. As calibrator sample cDNA from *orb2*$^+$ was used. Values of fold changes in *orb2*$^+$ sample versus *orb2*$^{\Delta A}$ or *orb2*$^{\Delta B}$ samples represent averages from three independent experiments each run in triplicates. Changes in gene expression were reported as fold changes relative to *orb2*$^+$. *P* values determined by one-sample t test for the null hypothesis that $\Delta\Delta$Ct equals $Orb2^+$ (H_0: LI = 1) or for unpaired t-test (H_0: $2^{-\Delta\Delta Ct}$*orb2*ΔA = $2^{-\Delta\Delta Ct}$ *orb2*ΔB).

Generation of the donor constructs containing modified genomic *orb2* fragments

***orb2*$^{+GFP}$** (in the main text also referred as *orb2*$^+$)

GFP tag has been PCR amplified with the primers SW11/SW12 and cloned as Asp718/BglII into the MCS of the vector containing attB- MCS- mFRT11- w+ (pSW11). 3'UTR of *orb2* was PCR amplified with SW15/SW16 primers and cloned downstream of the GFP as BglII/SpeI into pSW11. *orb2* ORF was PCR amplified with primers SW14/SW17 and cloned EcoRI/Asp718 in frame upstream of GFP, thus resulting in C-terminal fusion of *orb2* to GFP. The NheI site into the common exon after the splice site was inserted by single nucleotide exchange through overlap PCR using the primers SK001 and SK002. Resulting pKS12 vector was used for all subsequent cloning.

***orb2*$^{\Delta BGFP}$** (in the main text referred also as *orb2*$^{\Delta B}$)
The *orb2*$^{\Delta B}$ allele was generated by overlap PCR obtained from amplification of pSK12 using the primers SW14/SK054 and SK053/SK055 and subsequently replaced the pSK12 EcoRI/NheI fragment.

***orb2*$^{\Delta AGFP}$** (in the main text referred also as *orb2*$^{\Delta A}$)
The *orb2*$^{\Delta A}$ allele was generated by overlap PCR obtained from amplification of pSK12 using the primers SW14/SK054 and SK053/SK001 and subsequently replaced the pSK12 EcoRI/NheI fragment.

***orb2*$^{\Delta QGFP}$** (in the main text referred also as *orb2*$^{\Delta Q}$)
The *orb2*$^{\Delta Q}$ allele was generated by PCR amplification of the common *orb2* exon with the primers SK062 and SW17 and subsequent cloning into pSK12 with NheI/Asp718, deleting AA168-221 of *orb2*.

***orb2*^{RBD*GFP}**
To obtain point mutations in RRM1 (AA448-510), the common *orb2* exon was PCR amplified with the primers SK11/CP149 and the primers CP148/SW17. The resulting PCR products were used for overlap PCR using the primers SK11 and SW17 and subsequently cloned into pSK12 with NheI/Asp718 mutating Y492A, F494A. To obtain additional point mutations in RRM2 (AA556-634) the above construct with mutations in RRM1 was PCR amplified with the primers SK11/CP151 and the primers CP150/SW17. The resulting PCR products were used for overlap PCR using the primers SK11 and SW17 and subsequently cloned into pSK12 with NheI/Asp718 mutating Y492A, F494A, R601A, F604A.

Isoform specific deletion of the Q-domain and RBD* modifications
To obtain isoform specifc deletion of Q-domain and RBD* mutants the respective mutant construct was subcloned NheI/Asp718 into the respective isoform specific mutant.

***orb2*^{orb1RBDGFP}** (in the main text referred also as *orb2* ^{orb1RBD})
orb1 RBD was amplified from the genomic DNA from *Drosophila* w1118 using primers SK015 and SK006. The common *orb2* region was PCR amplified with the primers SK11 and SK14. The resulting PCR products were used for overlap PCR using the primers SK11 and SK006 and subsequently cloned into pSK12 with NheI/Asp718.

***orb2*^{mCPEB2RBDGFP}** (in the main text referred also as *orb2* ^{mCPEB2RBD})
mCPEB2 RBD was amplified from the mouse cDNA derived from brain tissue using the primers SK073 and SK071. The common *orb2* region was PCR amplified with the primers SK11 and SK072. The resulting PCR products were used for overlap PCR using the primers SK11 and SK071 and subsequently cloned into pSK12 with NheI/Asp718.

***orb2*^{orb1QGFP}** (in the main text referred also as *orb2* ^{orb1Q})
The Q-domain of *orb1* was amplified from the genomic DNA of *Drosophila* w1118 using primers SK003 and SK004. The common *orb2* region was PCR amplified with the

primers SK005 and SW17. The resulting PCR products were used for overlap PCR using the primers SK003 and SW17 and subsequently cloned into pSK12 with NheI/Asp718.

***orb2*$^{\textbf{mCPEB3QGFP}}$** (in the main text referred also as *orb2* $^{\text{mCPEB3Q}}$)
The Q-domain of *mCPEB3* was amplified from the *mouse* derived brain cDNA using primers SK007 and SK016. The common *orb2* region was PCR amplified with the primers SK017 and SW17. The resulting PCR products were used for overlap PCR using the primers SK007 and SW17 and subsequently cloned into pSK12 with NheI/Asp718.

***orb2*$^{\textbf{Scure2QGFP}}$** (in the main text referred also as *orb2* $^{\text{Scure2Q}}$)
The Q-domain of *Scure2* was amplified from *yeast c*DNA using primers SK029 and SK030. The common *orb2* region was PCR amplified with the primers SK031 and SW17. The resulting PCR products were used for overlap PCR using the primers SK029 and SW17 and subsequently cloned into pSK12 with NheI/Asp718.

***orb2*$^{\textbf{AcCPEBQGFP}}$** (in the main text referred also as *orb2* $^{\text{AcCPEBQ}}$)
The Q-domain of *AcCPEB* was amplified from Aplysia cDNA (gift of K.Si) using primers SK036 and SK040. The common *orb2* region was PCR amplified with the primers SK041 and SW17. The resulting PCR products were used for overlap PCR using the primers SK036 and SW17 and subsequently cloned into pSK12 with NheI/Asp718.

UAS-orb2B
orb2B was amplified from *Drosophila* w1118 cDNA using primers SK106 and SK107 and subsequently cloned into pKC26 (unpublished, map on the VDRC website) with EcoRI/BglII.

UAS-orb2BRBD*
To obtain point mutations in RRM1 (AA448-510), *orb2B* was PCR amplified from *Drosophila* w1118 cDNA with the primers SK106/CP149 and the primers CP148/SK107. The resulting PCR products were used for overlap PCR using the primers SK106 and SK107 and subsequently cloned into pKC26 (unpublished, map on the VDRC website) with EcoRI/BglII mutating Y492A, F494A. Donor constructs were injected into the fly strain bearing the acceptor site attP (Vie260b, VDRC) and phiC31 recombinase.

Primers used in this study

A1GCGCAGATCTCTAGGGCAGCAGCGGAACTGAGG
A2GCATGCGGCCGCACCATCATCGAAACGAGAGGTTTG
B1GCGCGGTACCTTGAGTGTGCAGGGGCTGTCCTTA
B2GCGCACTAGTGCGTAAAATTGAAATTGTTTCCACATCGTTC
b-GGAACTTAAGGACAGCCCCTGCACA
c-GACTATTTTGTGTCGCCTCGCTTTCG
e-ATGTACAACAAATTTGTTAATTTC
f-GTGTCGGTATATTCATGTGC
SB1TGATCGCCTCGAAAGCCAATCCTAC
SB2TTGAAAATCGGACAGCCAAATGCTC
SK001GTTGAGATTGAGATTCGGCAGGCTAGCGCAACTGCAGGAAGAAGGGACAAC
SK002GTTGTCCCTTCTTCCTGCAGTTGCGCTAGCCTGCCGAATCTCAATCTCAAC
SK003GCGCGCGCGGCTAGCTCTGTAAATAGCAACAAAATTCCACATC

SK004CTATGTGATCCTCCGCTTTCCCGAAGTTGTTGGCTACCCAAATTGGCAGATATC
SK005GATATCTGCCAATTTGGGTAGCCAACAACTTCGGGAAAGCGGAGGATCACATAG
SK006GCGCGGTACCGATCGCCTGATGCTGTTGACGCTGTATTC
Sk007GCGCGCGCGGCTAGCATGGACAAAAGCAAAACCCAGCCCCAG
SK011GCGCGCGCGCTAGCCTGCCGAATCTCAATCTCAAC
SK014GGAAGACCTTGGGAGAATAGTTGAGCATGCCATCGCCGACGTTTCCTCCATTTCC
SK015GGAAATGGAGGAAACGTCGGCGATGGCATGCTCAACTATTCTCCCAAGGTCTTCC
Sk016CTATGTGATCCTCCGCTTTCCCGAAGCTCGGGCTGGAGCTGTTGCTGC
SK017GCAGCAACAGCTCCAGCCCGAGCTTCGGGAAAGCGGAGGATCACATAG
SK029GCGCGCCGCGCTAGCAATAACAACGGCAACCAAGTGTCG
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CP148
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CP151
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HSC70fwCTCCTGCTGTTGGTATTGATTTGG
HSC70revTACGATTACCCTGGTCGTTGG
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Orb2ArevTTGTTGATGCTGTTGCTGGT
Orb2BfwGCATAGTCCCAGTGGAGGAG
Orb2BrevCAATCCCAGCCCGTTGTT
Orb2commonfwGCCTGCCGAATCTCAATCT

Orb2commonrevTGGTGCTGCTGATGCTGT

Supplemental References

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