

Figure S1

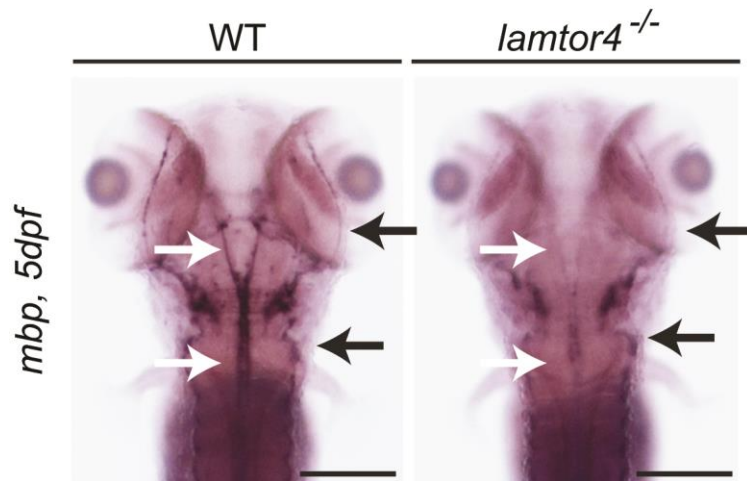


Figure S1. Related to Figure 1. *lamtor4* is essential for CNS myelination.

(A) Analysis of *mbp* mRNA expression at 5 dpf by whole mount *in situ* hybridization. Compared to their wildtype siblings, *lamtor4*^{-/-} mutants show reduced *mbp* expression in the CNS (white arrows), whereas *mbp* expression in the PNS is normal (black arrows). All panels show dorsal views, with anterior to the top. Genotypes were determined by PCR after imaging. Scale bar = 50 μ m.

Figure S2

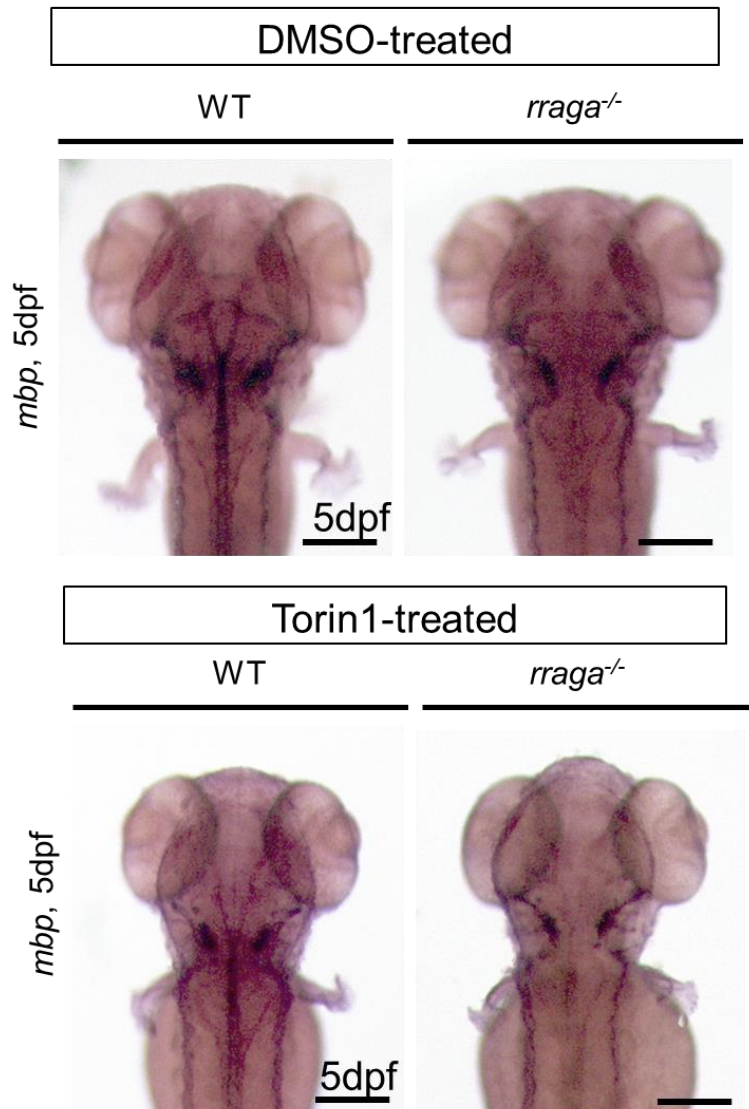


Figure S2. Related to Figure 1. Torin treated fish are developmentally delayed
mbp mRNA expression at 5 dpf as detected by whole mount *in situ* hybridization. Wildtype larvae treated with the mTOR signaling pathway inhibitor TORIN1 have delayed developmental progression and slightly reduced levels of *mbp* mRNA both in the CNS and PNS. Genotypes were determined by PCR after imaging. Scale bar = 50 μm.

Figure S3

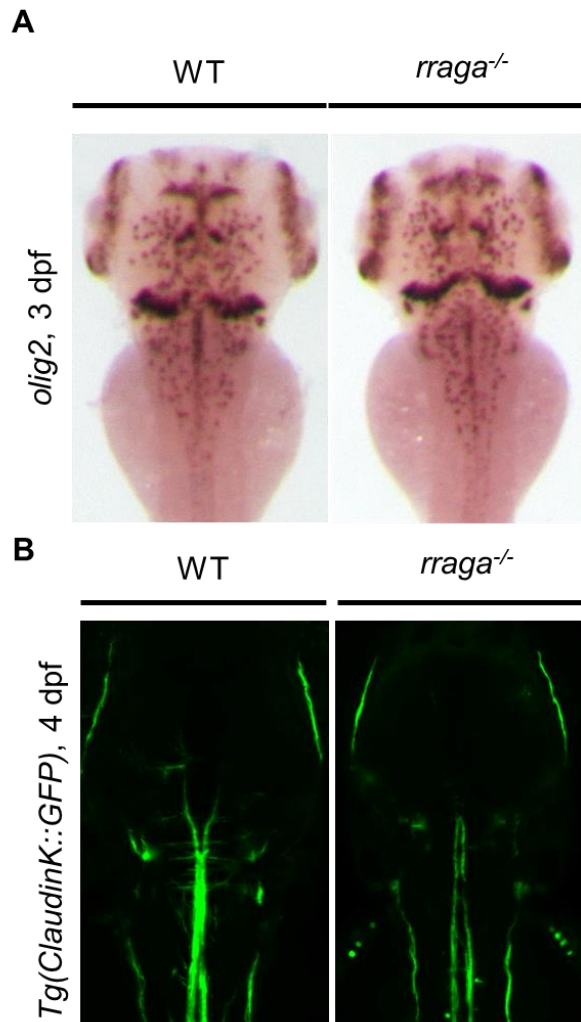


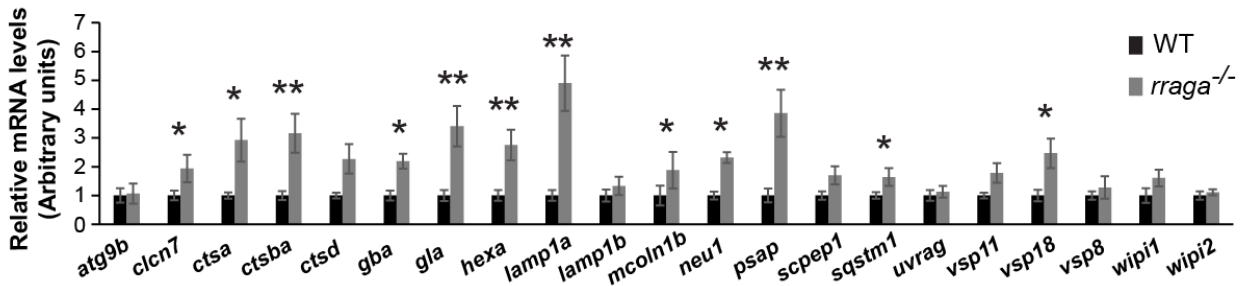
Figure S3. Related to Figure 2. *rraga*^{-/-} mutants have normal number of cells expressing *olig2*.

(A) Analysis of *olig2* mRNA expression at 3 dpf by whole mount *in situ* hybridization. Number of *olig2* positive cells is similar in *rraga*^{-/-} mutants and wildtype siblings.

(B) Dorsal view of 4 dpf *Tg(claudinK::GFP)* zebrafish larvae shows that *rraga*^{-/-} mutants express claudinK:GFP, but at reduced levels. Genotypes were determined by PCR after imaging

Figure S4

A



B

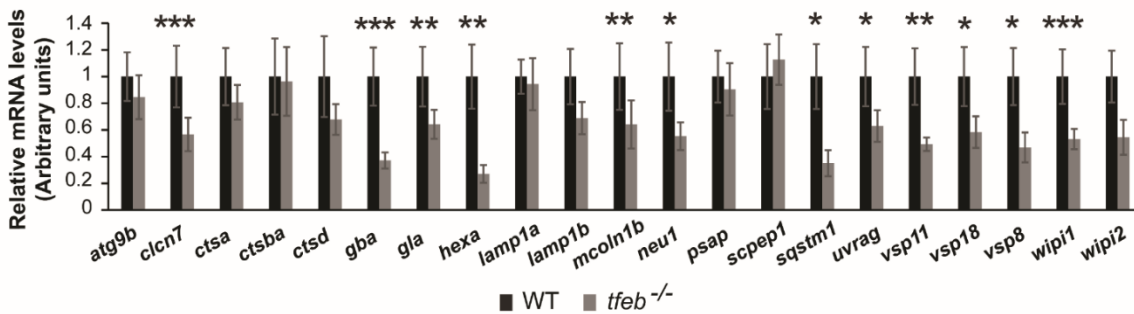


Figure S4. Related to Figure 3. RagA promotes CNS myelination by repressing TFEB

(A) Whole animal quantitative RT-PCR analysis reveals overexpression of some TFEB target genes in *rraga*^{-/-} mutants. (B) Whole animal qRT-PCR reveals reduced expression of some TFEB target genes in *tfeb*^{-/-} mutants. Graphs depict average values and standard error of the mean (** p < 0.01, Student T-test, two-tailed).

Figure S5

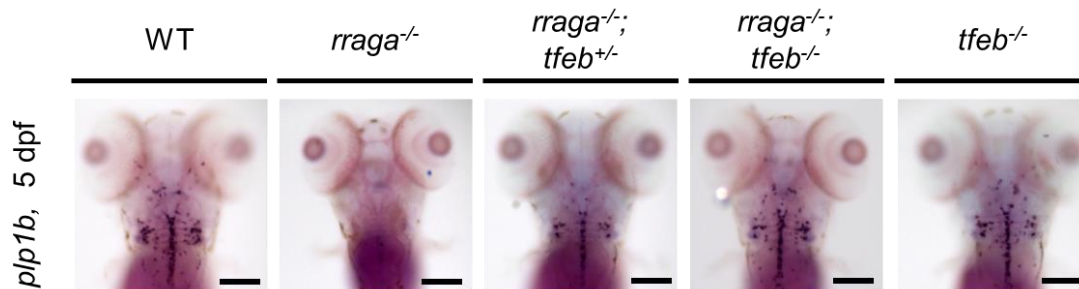


Figure S5. Related to Figure 3. *plp* expression is restored in *rraga*^{-/-};*tfeb*^{-/-} double mutants

Dorsal view of 5 dpf whole mount in situ hybridization to detect *plp1b* mRNA. Expression of *plp1b* mRNA is nearly absent in *rraga*^{-/-} mutants, but is restored in *rraga*^{-/-};*tfeb*^{-/-} double mutants. By this assay *rraga*^{-/-};*tfeb*^{-/-} double mutants and *tfeb*^{-/-} mutants are indistinguishable from wildtype larvae. Expression of *mbp* is also partly rescued in *rraga*^{-/-};*tfeb*^{+/-} mutants. Genotypes were determined by PCR after imaging. Scale bar = 50 μ m

Table S3

Table S3. Related to Figure 3. Upregulated or downregulated TFEB target genes in FAC-sorted *cldnk:GFP* positive cells of *rraga*^{-/-} mutants

Upregulated TFEB target genes		Downregulated TFEB target genes	
Zebrafish Gene	Human Gene	Zebrafish Gene	Human Gene
agtrap	AGTRAP	hoxc13a	HOXC13
amdhd2	AMDHD2	prkag2b	PRKAG2
atp6ap1b	ATP6AP1	naglu	NAGLU
atp6v0e1	atp6v0e1	comtd1	COMTD1
atp6v1ab	ATP6V1A		
cst14a.2	CSTB		
cst14b.1	CSTB		
ctsa	CTSA		
ctsba	CSTB		
flcn	FLCN		
hexb	HEXB		
hoxb7a	HOXB7		
ifi30	IFI30		
megf9	MEGF9		
sqstm1	SQSTM1		
tpp1	TPP1		
vps33a	VPS33A		