Supplemental Material for

Alternative Polyadenylation and miR-34 Family Members Regulate Tau Expression†

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Supplementary Table S1. Primers for 3'-RACE.

Purpose	*Location*	*Orientation*	*Sequence (5' to 3')*
3'-RACE Reverse Transcription Adaptor	Poly(A) Tail	Reverse	GCGAGCACAGAATTAATACGACTCACTATAGG-$(T)_{12}$-VN
3'-RACE Outer Primer	3'-RACE Adaptor	Reverse	GCTGATGGCGATGAATGAACACTG
Short Tau 3'-UTR Primer	Tau 3'-UTR 112-134	Forward	TCGGTTAATTGGTTAATCACTTA
Long Tau 3'-UTR Primer	Tau 3'-UTR 3531-3550	Forward	CCACTGACAGGCTTTCCCAG

Supplementary Table S2. Primers for Cloning.

Construct	*Tau 3'-UTR Location*	*Orientation*	*Sequence (5' to 3')*
Tau3UTR_WT	1-15	Forward	GCTCGCTAGCCTCGAGTCAGGCCCCTGGGGC
Tau3UTR_WT	4451-4465	Reverse	ATGCCTGCAGGTCGACTGGTGCCGGTCTTGC
Tau3UTR_short	1-15	Forward	GCTCGCTAGCCTCGAGTCAGGCCCCTGGGGC
Tau3UTR_short	223-256	Reverse	ATGCCTGCAGGTCGACTGAATGTTTTTTTTTAAA TATTTTATTACTAGCC
1-960	1-15	Forward	GCTCGCTAGCCTCGAGTCAGGCCCCTGGGGC
1-960	939-960	Reverse	ATGCCTGCAGGTCGACGGCAGCAGTTCCAACCTC AGA
954-1869	954-975	Forward	GCTCGCTAGCCTCGAGTGCTGCCATGATTTTGGC CACT
954-1869	1846-1869	Reverse	ATGCCTGCAGGTCGACTCCAACTACAACTCAACA GGGTGC
1845-2793	1845-1868	Forward	GCTCGCTAGCCTCGAGTGCACCCTGTTGAGTTGT AGTTGG
1845-2793	2774-2793	Reverse	ATGCCTGCAGGTCGACAGCTGCACACGAAGCTG CCA
2770-3729	2770-2798	Forward	GCTCGCTAGCCTCGAGGGGCTGGCAGCTTCGTGT GC
2770-3729	3709-3729	Reverse	ATGCCTGCAGGTCGACGGAGCCAGTGTGAGAGG TGGC
3730-4163	3730-3747	Forward	GCTCGCTAGCCTCGAGAGACACACAGCCTGTGCT
3730-4163	4133-4163	Reverse	ATGCCTGCAGGTCGACTAATCAGAGTAATAACTT TATTTCCAAATTC
954-1136	954-975	Forward	ATGCCTGCAGGTCGACTCCAACTACAACTCAACA GGGTGC
954-1136	1117-1136	Reverse	ATGCCTGCAGGTCGACGCAGTGGCCGTGGGAAG GAC
1114-1279	1114-1133	Forward	GCTCGCTAGCCTCGAGCCTGTCCTTCCCACGGCC AC
1114-1279	1259-1279	Reverse	ATGCCTGCAGGTCGACACCTCCTGCAACCAACCA GGG
1259-1869	1259-1279	Forward	GCTCGCTAGCCTCGAGCCCTGGTTGGTTGCAGGA GG
1259-1869	1846-1869	Reverse	ATGCCTGCAGGTCGACTCCAACTACAACTCAACA GGGTGC
2770-3259	2770-2798	Forward	GCTCGCTAGCCTCGAGGGGCTGGCAGCTTCGTGT GC
2770-3259	3237-3259	Reverse	ATGCCTGCAGGTCGACCCTATCTAGCCCACCCAA GGACA
3232-3501	3232-3251	Forward	GCTCGCTAGCCTCGAGGCCAGTGTCCTTGGGTGG GC
3232-3501	3482-3501	Reverse	ATGCCTGCAGGTCGACCGTGGCTGCTCCCTCCCT CT
3499-3729	3499-3520	Forward	GCTCGCTAGCCTCGAGACGGAGTTAGAGGCCCTT GGGG
3499-3729	3709-3729	Reverse	ATGCCTGCAGGTCGACGGAGCCAGTGTGAGAGG TGGC

Supplementary Table S3. Primers for Site-Directed Mutagenesis.

Construct	*Orientation*	*Sequence (5' to 3')*
Tau3UTR_long	Forward	TTTCCAAATTGATGGGTGGGCTAGTAATCAAATATTTAAAAAA AAACATTCAAAAACA
Tau3UTR_long	Reverse	TGTTTTTGAATGTTTTTTTTTAAATATTTGATTACTAGCCCACC CATCAATTTGGAAA
954-1869_mut	Forward	GTTGTCTGCCGTGAGAGCCCAATGTGACCCTATACCCCTCATC ACA
954-1869_mut	Reverse	TGTGATGAGGGGTATAGGGTCACATTGGGCTCTCACGGCAGA CAAC
3730-4163_mut	Forward	GTGTATTGTGTGTTTTAACAAATGATTTACACTCTGACAAGCT GTAAAAGTGAATTTGG
3730-4163_mut	Reverse	CCAAATTCACTTTTACAGCTTGTCAGAGTGTAAATCATTTGTT AAAACACACAATACAC

Supplementary Table S4. miRNA-Related Oligonucleotides.

Oligonucleotide	*Sequence (5' to 3')*
Pre-miR Negative Control #1	Proprietary (Life Technologies Catalog # AM17110)
Pre-miR-34a-5p	Proprietary (Life Technologies Catalog # PM11030)
Pre-miR-132-3p	Proprietary (Life Technologies Catalog # PM10166)
Pre-miR-181c-5p	Proprietary (Life Technologies Catalog # PM10181)
Pre-miR-642-5p	Proprietary (Life Technologies Catalog # PM11477)
miRCURY LNA Negative Control A	GTGTAACACGTCTATACGCCCA
miRCURY LNA hsa-miR-34a-5p	ACAACCAGCTAAGACACTGCC
miRCURY LNA hsa-miR-34b-5p	AATCAGCTAATGACACTGCCT
miRCURY LNA hsa-miR-34c-5p	GCAATCAGCTAACTACACTGCC

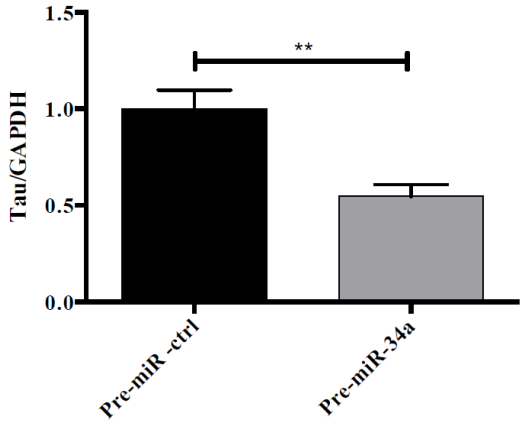
Supplementary Table S5. Custom Standards for Absolute Quantification qPCR.

Gene	*Sequence*
Tau	GTAAAATCTGAGAAGCTTGACTTCAAGGACAGAGTCCAGTCGAAGATTGGGTCCC TGGACAATATCACCCACGTCCCTGGCGGAGGAAATAAAAAGATTGAAACCCACA AGCTGACCTTCCGCGAGAACGCCAAAGCCAAGACAGACCACGGGGCGGAGATCG TGTACAAGTCGCCAGTGGTGTCTGGGGACACGTCTCC
GAPDH	AGCCCGCAGCCTCCCGCTTCGCTCTCTGCTCCTCCTGTTCGACAGTCAGCCGCATC TTCTTTTGCGTCGCCAGCCGAGCCACATCGCTCAGACACCATGGGGAAGGTGAAG GTCGGAGTCAACGGATTTGGTCGTATTGGGCGCCTGGTCACCAGGGCTGCTTTTA ACTCTGGTAAAGTGGATATTGTTGCCATCAATGACCCCTTCATTGACCTCAACTAC ATGGTTTACATGTTCCAATATGATTCCACCCATGGCAAATTCCATGGCACCGTCAA GGCTGAGAACGGGAAGCTTGTC

Supplementary Table S6. TaqMan Gene Expression Assay Probes.

Target	*Oligonucleotide Type*	*Sequence (5' to 3')*
Firefly luciferase (*luc2*)	Forward Primer	GCGCAGCTTGCAAGACTATAAG
	Reverse Primer	TTGTCGATGAGAGTGCTCTTAGC
	TaqMan Probe	CTGGTGCCCACACTAT
Renilla luciferase *(hRluc*)	Forward Primer	CCTCCTGGATCACTACAAGTACCT
	Reverse Primer	GTGGCCCACAAAGATGATTTTCT
	TaqMan Probe	CAGCAGCTCGAACCAA
Tau	TaqMan Assay	Proprietary (Life Technologies Assay ID: Hs00902194_m1)
GAPDH	TaqMan Assay	Proprietary (Life Technologies Assay ID: Hs99999905_m1)

Supplementary Figure S1. Quantification by densitometry of western blot shown in Figure 6A combined with that of a duplicate experiment (n = 3 technical replicates times n = 2 biological replicates).



Supplementary Figure S2. Quantification by densitometry of western blot from Figure 6B, transfected with Lipofectamine 2000 (A) and that of a similar experiment transfected using RNAiMAX (B). n = 3 technical replicates for each experiment. While neither reached statistical significant, note the trend in both experiments toward increased tau protein levels upon treatment with LNA targeting miR-34a/b/c.