

**Table S1.** Cohort demographics

	AD patients	Non-demented controls
N	12	12
Sex	Female	Female
Age of death; Mean (SD)	84.5 (5.04)	77.92 (13.00)
PMI; Mean (SD)	2.89 (0.96)	2.48 (0.62)
Plaque total; Mean (SD)	13.69 (1.71)	2.16 (3.54)
Tangle total; Mean (SD)	14.0 (1.36)	3.33 (2.38)
Braak stage; Range	V-VI	I – IV

Dorsal raphe nucleus (DRN) tissue was obtained from the Banner Sun Health Research Institute (BSHRI, Sun City, Arizona, US) as part of the Brain and Body Donation Program (BBDP). Displayed is the number of samples in each group, the sex of the subjects and the mean plus standard deviation (SD) of their age of death, post-mortem interval (PMI, in hours), total plaque load, and total tangle load, as well as their range of Braak stage.

**Table S2.** Primer overview

PCR primers				
Gene	Forward primer (5'-3')	Reverse primer (5'-3')	Target region (GRCh37)	Product size (bp)
RHBDF2 (Multiplex)	AGGGTTTTGGGGATTTAG TG	CCAAAAACCTCACAAAC AAATC	17:74475178 -74475497	320
RHBDF2 (Singleplex)	AGGGTTTTGGGGATTTAG TG	(Bio-)CATCACCCCTTCCACAC ACTCAA	17:74475178 -74475435	258
OXT (Multiplex)	GTTTAGGTTTTGTTAATG AAGAGGAA	ACCAAACTAAAACTC ACCTTAC	20:3052117-3052440	324
OXT (Singleplex)	GTTTAGGTTTTGTTAATG AAGAGGAA	(Bio-)TCTTACCTCCCAAAAAA CAATTC	20:3052117-3052392	276
TNXB (Multiplex)	TGTGAAGTAGGTTATTTA GGGGAAGA	CCTCCAAAACAAACCCTA ACTAAAACTCT	6:32063558-32063997	440
TNXB (Singleplex)	TGTGAAGTAGGTTATTTA GGGGAAGA	(Bio-)ACAAACCCTAACTAAAA ACTCTTCCTACC	6:32063566-32063997	432
DNAJB13 (Multiplex)	GGTATTTTGGGAGGATGA GTTA	CCCTTCAAAAACAAACC AACTAAT	11:73668561 -73668825	265
DNAJB13 (Singleplex)	GTATTTTGGGAGGATGAG TTATAATTG	(Bio-)CTTCAAAAACAAACCAA CTAATAC	11:73668563 -73668824	262
PGLYRP1 (Multiplex)	GTTGGGGAGAGTTTAGGT AAG	ATTCTATTAAATTATTCA ATATTCCACT	19:46526238 -46526634	397
PGLYRP1 (Singleplex)	TGGGGAGAGTTTAGGTA AGTA	(Bio-)TCCTATTAAATTATTCAA TATTCCACTAA	19:46526240 -46526632	393
C3 (Multiplex)	GATTGGGTTTTATTTGAG TGTAAGAT	CCAACAATAAATTAAAA ACTCCAATCTTC	19:6713187-6713627	441

C3 (Singleplex)	GATTGGGTTTTATTTGAG TGTAAGAT	(Bio- )AACCTAAAACCCTCCTT ATCTAT	19:6713187- 364 6713550
LMX1B (Multiplex)	TTTTTTAAGGGGGTGGAG TAGAG	CCTCACCTCCCCAAACTT CCTAACATT	9:129375928 257 -129376184
LMX1B (Singleplex)	GGGGGTGGAGTAGAGGG A	(Bio- )CCTCACCTCCCCAAACTT CCTAACATT	9:129375936 249 -129376184

#### Pyrosequencing primers

Gene	Sequencing primer (5'-3')	Number of CpGs	PyroMark Orientation	Target region (GRCh37)	Primer
RHBDF2	TTTTTGGTTGGGGAG	5	Upper strand(5'-3')	17:74475230-74475271	1
RHBDF2	GTATGTTGTAGTGGTG	5	Upper strand(5'-3')	17:74475287-74475332	2
RHBDF2	GATTTTAAATTATTGGGTTG	4	Upper strand(5'-3')	17:74475355-74475403	3
OXT	GGTTTTGTTAATGAAGAGGA A	5	Upper strand (5'-3')	20:3052143-3052173	1
OXT	ATATTATTAATTTTTTAAATA GAG	2	Upper strand (5'-3')	20:3052215-3052224	2
OXT	AATGTTTAGGTATAAAAAGG T	5	Upper strand (5'-3')	20:3052248-3052297	3
TNXB	GGTTAGTGTTTAGATGGG	5	Lower strand (5'-3')	6:32063874-32063913	1
TNXB	AGGATTGAGGTGTGA	4	Lower strand (5'-3')	6:32063823-32063867	2
TNXB	GGTGTGTGTATTTGTTG	2	Lower strand (5'-3')	6:32063774-32063806	3
DNAJB13	GGATGAGTTATAATTGGG	5	Lower strand (5'-3')	11:73668745-73668795	1
DNAJB13	TTTGTATTGTTTGAAAA	4	Lower strand (5'-3')	11:73668618-73668673	2
PGLYRP1	TTTTTTTTTGGTTGGGTTAG	1	Upper strand (5'-3')	19:46526451-46526454	1
PGLYRP1	GAGGGATGTATTGTGG	3	Upper strand (5'-3')	19:46526468-46526509	2
PGLYRP1	GTTTAGTAGGGAGGG	5	Upper strand (5'-3')	19:46526524-46526555	3
C3	TTTAAATTTGAGAAGGGAGA	2	Upper strand (5'-3')	19:6713340-6713376	1
C3	TGGGGTAGGGATTAAA	4	Upper strand (5'-3')	19:6713398-6713426	2
C3	ATTTAGGTAGGGAAA	2	Upper strand (5'-3')	19:6713450-6713475	3
LMX1B	GTTTAGAAGAAGATTAAAAT TTTTG	7	Upper strand (5'-3')	9:129375982-129376016	1

Displayed is an overview of the polymerase chain reaction (PCR) and pyrosequencing primers that were used in this study. Provided for each target gene are the multiplex and singleplex primer sequences (Bio, biotinylated), as well as corresponding genomic coordinates (Ensembl GRCh37 assembly) and amplicon sizes in base pairs

(bp). For the pyrosequencing primers, the total number of CpG sites, the targeted region for sequencing, and the orientation of the assay in the PyroMark assay design software 2.0 are listed per gene.

**Figure S1.** Allele overview. Displayed for each target gene are the recovered alleles and their respective methylation patterns. Each horizontal row represents a single product-yielding polymerase chain reaction (PCR) containing either one, two or three alleles, indicated with 1, 2 or 3, respectively. Cytosine-phosphate-guanine (CpG) sites are depicted as boxes and colored according to their methylation pattern. Please refer to the figure legend for further details on color-coding.

