

## Supplemental Information

**Supplemental Figure 1. A.** IB of the wild type (WT) mouse brain tissue, the kloθο hypomorph (*kl/kl*) brain tissue, and the spK11 transiently transfected cell lysate, representing the positive control for the 70 kDa band for spliced KL1, using anti-KL1 and anti-spE15. The anti-KL1 IB shows a 130 kDa band in the WT brain which is not present in the kloθο hypomorph (*kl/kl*) brain. The presence of 2 lower bands, ~100 kDa and ~80 kDa are visible in both the WT and *kl/kl* brain tissue, which renders them to be “non-kloθο” protein bands. **B.** IP and IB of the WT and *kl/kl* mouse brain tissue, using our SB48 synthetic anti- $\alpha$ Kloθο<sup>53</sup> on top and anti-KL1 at the bottom, for IP of KL1. The IB was performed using KM2076 (anti-KL1) and anti-spE15 respectively. In all four immunoblots, no full-length  $\alpha$ kloθο (130 kDa) or KL1 (~75 kDa) was precipitated from the kloθο hypomorph (*kl/kl*) brain tissue as compared to the WT brain tissue where both bands can be seen in the anti-KL1 IB. These positive Kloθο bands in the WT brain though, cannot be detected in the anti-spE15 IB, nor can any 70 kDa band be seen for the *kl/kl* brain. A fainter set of bands nearing 150 kDa can be seen in both WT and *kl/kl* in the IP: anti-KL1/IB: anti-spE15 combo, which are most-likely non-specific.

Supplemental Table 1:  $\alpha$ -Klotho PCR primers

<b>A. Human full-length transmembrane <math>\alpha</math>-Klotho</b>	5'- TGATAGGATCCGCCGCCACCATGCCCCGCCAGCGCCCCGCCGC-3' 5'-CGTCCTCTAGAGGTGTAAAAAACTGCAC-3'
<b>B. Human <math>\alpha</math>-Klotho peptides</b>	
Peptide A:	5'-TGATAGGATCCGCCGCCACCATGCCCCGCCAGCGCCCCGCCGC-3' 5'-CGTCCTCTAGAGGGGTTGTCGATGGTGAT-3'
Peptide B:	5'-TGATAGGATCCGCCGCCACCATGGAGGGGCTGCGCTACTA-3' 5'-CGTCCTCTAGAGTGAGGGTCCAAAAGTTG-3'
Peptide C:	5'-TGATAGGATCCGCCGCCACCATGATGACCGACCACAGCAT-3' 5'-CGTCCTCTAGATACTTGAATGTAGTTGTCAAC-3'
Peptide D:	5'-TGATAGGATCCGCCGCCACCATGGATACCACTCTGTCTCAG-3' 5'-CGTCCTCTAGAGGCCACCTCTTTGTCC-3'
Peptide E:	5'-TGATAGGATCCGCCGCCACCATGCTCGGCCATCACGTCAAGC-3' 5'-CGTCCTCTAGAGTACATGGGGAGGTCTC-3'
Peptide F:	5'-TGATAGGATCCGCCGCCACCATGCTAATCCAGGGTACCTTTG-3' 5'-CGTCCTCTAGAGGTGTGAAAAAACTGCAC-3'
<b>C. Mouse full-length soluble Klotho</b>	5'-TGATAGAATTCGCCGCCACCATGCTAGCCCGCGCCCCTCCT-3' 5'-CAGCTGCGGCCGCTCATTCAAGGGGCTGGTTTTTC-3'
<b>D. Mouse Splice K11</b>	5'-TGATAGAATTCGCCGCCACCATGCTAGCCCGCGCCCCTCCT-3' 5'- CAGCTTCTAGAGTGAGGAAGCAAGAGGCCGACACTGGGTTTTGTC AA AGGACTTACTTGAACGTAGTTGTCAACAACTCC-3'

**Supplemental table 2** Primers used for detection of klotho transcript.

Pf1, Pr1 for mKL:	Pf1 5' ATGCTAGCCCGCGCCC 3' Pr1 5' ATCCACATTCGGTGCAC 3'
Pf1, Pr2 for mKL1:	Pf1 5' ATGCTAGCCCGCGCCC 3' Pr2 5' GTTGTCAACAACCTCCCC 3'
Pf1, Pr3 for spKL1:	Pf1 5' ATGCTAGCCCGCGCCC 3' Pr3 5' TTAGTGAGGAAGCAAGA 3'
Pf2, Pf3, Pr3 for 50KDa protein (spKL1):	Pf2 5' TGGCAGGGGTATGCCAC 3' Pr3 5' TTAGTGAGGAAGCAAGA 3' Pf3 5' CTGGTTGCCACAACT 3' Pr3 5' TTAGTGAGGAAGCAAGA 3'