

Supplementary data to:

**LONG-TERM ADMINISTRATION OF MELATONIN ATTENUATES
 NEUROINFLAMMATION IN THE AGED MOUSE BRAIN**

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Supplementary Table 1: The effect of melatonin on CD11b and GFAP protein levels in the hippocampus and the prefrontal cortex of aged mice

Type of protein	Brain area	Number of animal	Animal group		
			Young adult mice	Aged mice	Melatonin-treated aged mice
CD11b	Hippocampus	N ₁	100	169.9	114.5
		N ₂	100	158.4	95.9
		N ₃	100	240.3	153.9
		N ₄	100	177.6	134.3
		Mean±S.E.M.	100	186.7±18.3**	124.7±12.5[#]
	Prefrontal cortex	N ₁	100	259.1	187.8
		N ₂	100	195.1	154.3
		N ₃	100	182.4	94.3
		N ₄	100	210.1	152.6
		Mean±S.E.M.	100	211.7±16.8**	147.3±19.4[#]
GFAP	Hippocampus	N ₁	100	171.6	101.6
		N ₂	100	208.3	170.0
		N ₃	100	272.4	150.2
		N ₄	100	194.8	116.4
		Mean±S.E.M.	100	211.8±21.6**	134.6±15.6[#]
	Prefrontal cortex	N ₁	100	232.6	176.8
		N ₂	100	281.4	210.4
		N ₃	100	429.9	213.2
		N ₄	100	395.9	156.1
		Mean±S.E.M.	100	335.0±46.6***	189.1±13.8[#]

CD11b and GFAP protein levels were determined by Western blot analyses. A one-way ANOVA was performed for statistical analysis. Data represent the mean ± S.E.M. from 4 mice. ** $p < 0.01$ and *** $p < 0.001$ compared with young adult mice, and [#] $p < 0.05$ compared with aged mice

Supplementary Table 2: The effect of melatonin on IL-1 β , IL-6, and TNF- α protein levels in the hippocampus and the prefrontal cortex of aged mice

Type of protein	Brain area	Number of animal	Animal group		
			Young adult mice	Aged mice	Melatonin-treated aged mice
IL-1 β	Hippocampus	N ₁	100	264.6	179.4
		N ₂	100	292.2	152.8
		N ₃	100	236.9	125.1
		N ₄	100	197.4	99.0
		Mean\pmS.E.M.	100	247.8\pm20.2^{***}	139.1\pm17.4^{##}
	Prefrontal cortex	N ₁	100	249.6	208.4
		N ₂	100	305.6	124.1
		N ₃	100	430.8	180.5
		N ₄	100	346.7	170.7
		Mean\pmS.E.M.	100	333.2\pm38.1^{***}	170.9\pm17.5^{##}
IL-6	Hippocampus	N ₁	100	165.3	111.9
		N ₂	100	126.9	104.1
		N ₃	100	182.8	143.8
		N ₄	100	147.6	123.7
		Mean\pmS.E.M.	100	155.7\pm12.0^{**}	120.9\pm8.6[#]
	Prefrontal cortex	N ₁	100	185.5	105.4
		N ₂	100	149.8	130.0
		N ₃	100	147.9	132.7
		N ₄	100	196.9	120.2
		Mean\pmS.E.M.	100	170.0\pm12.5^{***}	122.1\pm6.2^{##}
TNF- α	Hippocampus	N ₁	100	131.5	82.9
		N ₂	100	115.5	94.4
		N ₃	100	121.0	100.3
		N ₄	100	105.5	92.5
		Mean\pmS.E.M.	100	118.4\pm5.4[*]	92.5\pm3.6^{##}
	Prefrontal cortex	N ₁	100	108.4	83.6
		N ₂	100	135.0	93.0
		N ₃	100	120.3	80.5
		N ₄	100	117.6	75.8
		Mean\pmS.E.M.	100	120.3\pm5.5[*]	83.2\pm3.6^{###}

IL-1 β , IL-6, and TNF- α protein levels were determined by Western blot analyses. A one-way ANOVA was performed for statistical analysis. Data represent the mean \pm S.E.M. from 4 mice. * p < 0.05, ** p < 0.01 and *** p < 0.001 compared with young adult mice, and # p < 0.05, ## p < 0.01 and ### p < 0.001 compared with aged mice.

Supplementary Table 3: The effect of melatonin on pNF-κB protein levels in the hippocampus and the and the prefrontal cortex of aged mice

Type of protein	Brain area	Number of animal	Animal group		
			Young adult mice	Aged mice	Melatonin-treated aged mice
pNF-κB	Hippocampus	N ₁	100	131.0	88.0
		N ₂	100	150.1	110.3
		N ₃	100	140.4	86.7
		N ₄	100	124.0	78.5
		Mean±S.E.M.	100	136.4±5.7**	90.9±6.8###
	Prefrontal cortex	N ₁	100	113.5	79.0
		N ₂	100	170.4	82.7
		N ₃	100	192.2	121.2
		N ₄	100	122.5	84.0
		Mean±S.E.M.	100	149.7±18.9*	91.7±9.9#

pNF-κB protein levels were determined by Western blot analyses. A one-way ANOVA was performed for statistical analysis. Data represent the mean ± S.E.M. from 4 mice. * $p < 0.05$ and ** $p < 0.01$ compared with young adult mice, and # $p < 0.05$ and ### $p < 0.001$ compared with aged mice.

Supplementary Table 4: The effect of melatonin on NR2A, NR2B, and CaMKII protein levels in the hippocampus and the prefrontal cortex of aged mice

Type of protein	Brain area	Number of animal	Animal group		
			Young adult mice	Aged mice	Melatonin-treated aged mice
NR2A	Hippocampus	N ₁	100	76.1	98.9
		N ₂	100	53.3	66.6
		N ₃	100	70.7	95.5
		N ₄	100	64.4	89.5
		Mean±S.E.M.	100	66.1±4.9**	87.6±7.3[#]
	Prefrontal cortex	N ₁	100	78.2	120.5
		N ₂	100	80.2	101.5
		N ₃	100	61.7	91.2
		N ₄	100	77.9	81.7
		Mean±S.E.M.	100	74.5±4.3*	98.7±8.3[#]
NR2B	Hippocampus	N ₁	100	84.5	79.9
		N ₂	100	62.1	77.7
		N ₃	100	66.6	86.8
		N ₄	100	64.3	92.3
		Mean±S.E.M.	100	69.4±5.1***	84.2±3.3[#]
	Prefrontal cortex	N ₁	100	74.5	97.0
		N ₂	100	84.9	102.1
		N ₃	100	65.9	81.3
		N ₄	100	38.5	94.9
		Mean±S.E.M.	100	66.0±9.9*	93.8±4.4[#]
CaMKII	Hippocampus	N ₁	100	82.4	100.3
		N ₂	100	70.6	120.1
		N ₃	100	74.4	85.7
		N ₄	100	77.7	83.0
		Mean±S.E.M.	100	76.3±2.5*	97.3±8.5[#]
	Prefrontal cortex	N ₁	100	53.4	88.2
		N ₂	100	52.2	100.9
		N ₃	100	101.5	107.0
		N ₄	100	77.1	102.9
		Mean±S.E.M.	100	71.1±11.7*	99.8±4.1[#]

NR2A, NR2B, and CaMKII protein levels were determined by Western blot analyses. A one-way ANOVA was performed for statistical analysis. Data represent the mean ± S.E.M. from 4 mice. * $p < 0.05$, ** $p < 0.01$ and *** $p < 0.001$ compared with young adult mice, and [#] $p < 0.05$ compared with aged mice.

Supplementary Table 5: The effect of melatonin on BDNF protein levels in the hippocampus and the prefrontal cortex of aged mice

Type of protein	Brain area	Number of animal	Animal group		
			Young adult mice	Aged mice	Melatonin-treated aged mice
BDNF	Hippocampus	N ₁	100	54.6	73.8
		N ₂	100	73.5	108.2
		N ₃	100	71.8	113.4
		N ₄	100	77.1	82.7
		Mean±S.E.M.	100	69.3±5.0*	94.5±9.6[#]
	Prefrontal cortex	N ₁	100	54.9	95.7
		N ₂	100	85.6	84.2
		N ₃	100	74.1	87.5
		N ₄	100	79.5	92.4
		Mean±S.E.M.	100	73.5±6.6**	90.0±2.5[#]

BDNF protein levels were determined by Western blot analyses. A one-way ANOVA was performed for statistical analysis. Data represent the mean ± S.E.M. from 4 mice. * $p < 0.05$, ** $p < 0.01$ compared with young adult mice, and [#] $p < 0.05$ compared with aged mice