

Supplemental Material:

Brief Report: Tamoxifen application is associated with transiently increased loss of hippocampal neurons following virus infection

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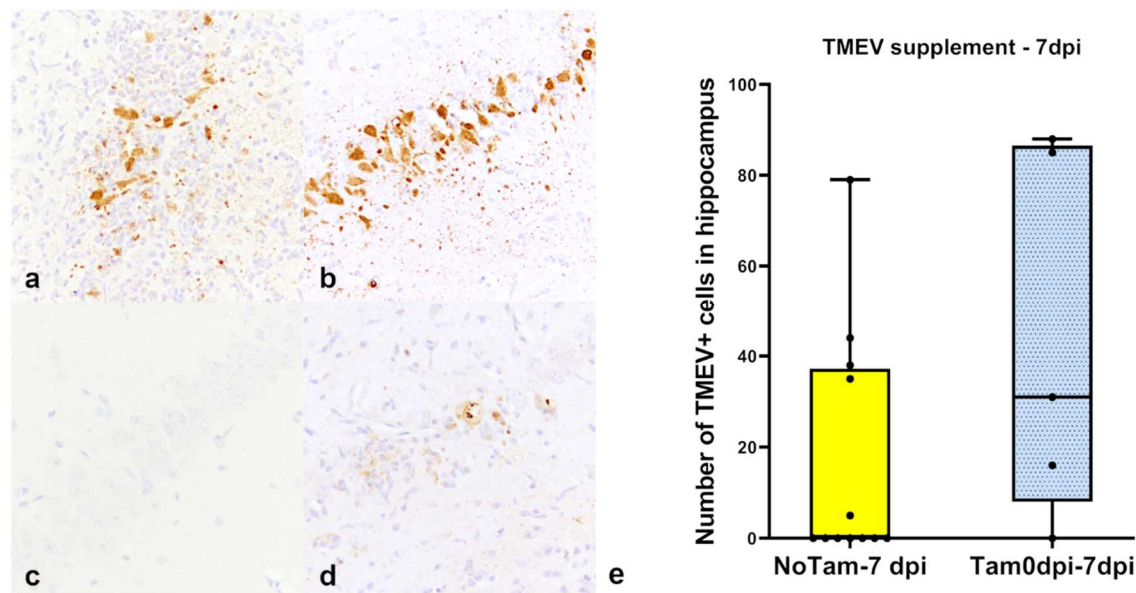
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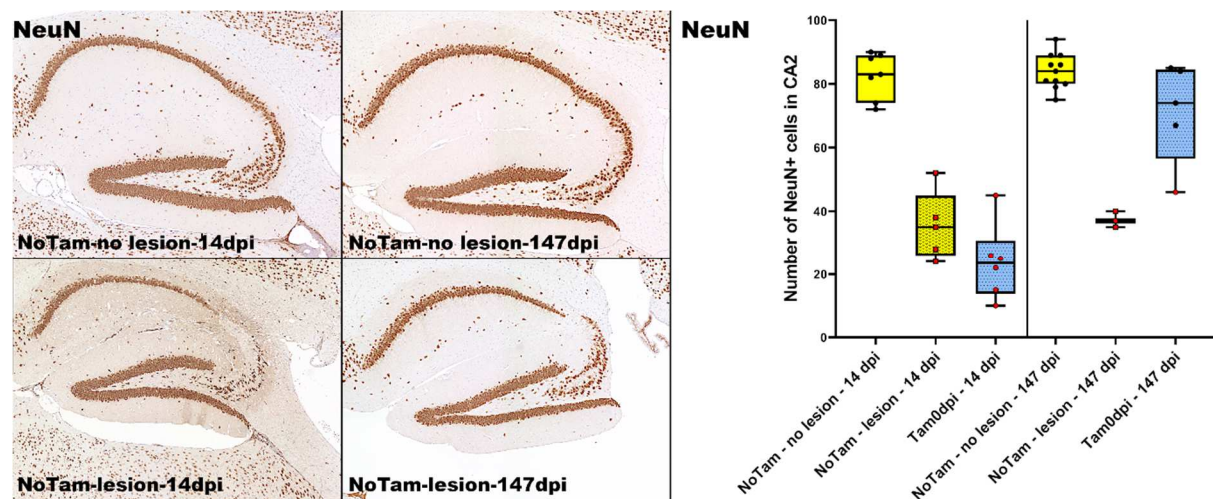
Supplementary Table S1: statistically significant p-values (Mann-Whitney U test) depicted in figure 1:

Antibody	Group 1	n=	vs. Group 2	n=	p-value
Amigo 2	NoTam-no lesion	7	Tam0dpi	6	0.005
Amigo 2	NoTam-no lesion	7	Tam5dpi	10	0.004
Amigo 2	NoTam-lesion	5	Tam5dpi	10	0.01
Aquaporin 4	NoTam-no lesion	7	NoTam-lesion	5	0.005
Aquaporin 4	NoTam-no lesion	7	Tam0dpi	6	0.005
Aquaporin 4	NoTam-no lesion	7	Tam5dpi	10	0.0002
CD3	NoTam-no lesion	7	NoTam-lesion	5	0.003
CD3	NoTam-no lesion	7	Tam0dpi	6	0.001
CD3	NoTam-no lesion	7	Tam5dpi	10	0.0007
CD3	Tam0dpi	6	Tam5dpi	10	0.007
DCX	NoTam-no lesion	7	Tam0dpi	6	0.001
DCX	Tam0dpi	6	Tam5dpi	10	0.002
GFAP	NoTam-no lesion	7	NoTam-lesion	5	0.005
GFAP	NoTam-no lesion	7	Tam0dpi	6	0.001
GFAP	NoTam-no lesion	7	Tam5dpi	10	0.0001
GFAP	NoTam-lesion	5	Tam0dpi	6	0.009
GFAP	NoTam-lesion	5	Tam5dpi	10	0.0007
GFAP	Tam0dpi	6	Tam5dpi	10	0.001
IBA-1	NoTam-no lesion	7	NoTam-lesion	5	0.005
IBA-1	NoTam-no lesion	7	Tam0dpi	6	0.001
IBA-1	NoTam-no lesion	7	Tam5dpi	10	0.01
Nestin	NoTam-no lesion	7	NoTam-lesion	5	0.003
Nestin	NoTam-no lesion	7	Tam0dpi	6	0.001
Nestin	NoTam-no lesion	7	Tam5dpi	10	0.0001
Nestin	NoTam-lesion	5	Tam0dpi	6	0.004
Nestin	NoTam-lesion	5	Tam5dpi	10	0.04
NeuN	NoTam-no lesion	7	NoTam-lesion	5	0.003
NeuN	NoTam-no lesion	7	Tam0dpi	6	0.001
NeuN	NoTam-no lesion	7	Tam5dpi	10	0.0002
NeuN	NoTam-lesion	5	Tam0dpi	6	0.02
S100A10	NoTam-no lesion	7	Tam0dpi	6	0.05
S100A10	NoTam-no lesion	7	Tam5dpi	10	0.007
TMEV	NoTam-no lesion	7	Tam0dpi	6	0.05
TMEV	NoTam-no lesion	7	Tam5dpi	10	0.009
TMEV	NoTam-lesion	5	Tam5dpi	10	0.02

ST1: Statistically significant p-values ($p \leq 0.05$) evaluated by Mann-Whitney U test presented in figure 1 of the main manuscript. Further information about the group labeling and composition can be found in table 1 of the main manuscript. Information about the used antibodies for immunohistochemistry can be found in the materials and methods section of the main manuscript. Significant p-values were depicted in the graphs of figure 1 of the main manuscript as asterisk (*).



Supplementary figure S1: Hippocampi of TMEV-BeAn infected mice at 7 dpi (a, b) and 14 dpi (c, d). Untreated mice (*NoTam-* a, c) and mice with tamoxifen application 0, 2 and 4 dpi (*Tam0dpi-* b, d) – immunohistochemistry, TMEV 400x magnification. Numerous TMEV antigen positive cells were present in both groups at 7 dpi (a,b) while only few positive cells were detectable in tamoxifen treated mice at 14 dpi (d), compared to untreated mice (c). (e) Number of TMEV antigen positive cells within the hippocampus at 7 dpi comparing untreated mice (*NoTam-* yellow, n=12) and mice with tamoxifen application 0, 2 and 4 dpi (*Tam0dpi-* light blue, dotted, n=5). There is no significant difference between the groups at 7 dpi ($p=0.148$) which is in contrast to the findings at 14 dpi (see main manuscript figure 1).



Supplementary figure S2: Hippocampi of untreated, TMEV-infected mice (*NoTam*) without (*no lesion*) and with neuronal loss (*lesion*) at 14 and 147 dpi – IHC NeuN. The graph depicts the untreated mice (yellow) without (*no filling*) and with neuronal loss (*v-pattern*) and mice with tamoxifen treatment (blue, dotted) at 14 and 147 dpi. Mice with neuronal loss are additionally marked with a red dot. 7 of 12 untreated mice (58 %) do not show neuronal loss at 14 dpi. 5 of 12 untreated mice (42%) and 6 of 6 mice (100%) with tamoxifen application at 0, 2 and 4 dpi (*Tam0dpi*) are affected by neuronal loss at 14 dpi. Most mice (79-80%) do not show neuronal loss at 147 dpi. 20-21 % of the mice (*NoTam*: 3 of 14; *Tam0dpi*: 1 of 5) still show neuronal loss at 147 dpi. Tamoxifen application had no long-term effect on the reduction of neuronal loss ($p=0.8$).